

This Page Is Inserted by IFW Operations
and is not a part of the Official Record

BEST AVAILABLE IMAGES

Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images may include (but are not limited to):

- BLACK BORDERS
- TEXT CUT OFF AT TOP, BOTTOM OR SIDES
- FADED TEXT
- ILLEGIBLE TEXT
- SKEWED/SLANTED IMAGES
- COLORED PHOTOS
- BLACK OR VERY BLACK AND WHITE DARK PHOTOS
- GRAY SCALE DOCUMENTS

IMAGES ARE BEST AVAILABLE COPY.

**As rescanning documents *will not* correct images,
please do not report the images to the
Image Problem Mailbox.**

(19) World Intellectual Property Organization
International Bureau



(43) International Publication Date
1 March 2001 (01.03.2001)

PCT

(10) International Publication Number
WO 01/14420 A2

(51) International Patent Classification⁷: C07K 14/00

the University of California, 12th floor, 1111 Franklin Street, Oakland, CA 94607-5200 (US). TAMAGNONE, Luca [IT/IT]; Corso Einaudi, 43, I-10129 Torino (IT).

(21) International Application Number: PCT/US00/23365

(22) International Filing Date: 25 August 2000 (25.08.2000)

(74) Agent: COX, Niki, D.; Biogen, Inc., 14 Cambridge Center, Cambridge, MA 02142 (US).

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
60/150,576 25 August 1999 (25.08.1999) US

(81) Designated States (*national*): AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW.

(71) Applicants (*for all designated States except US*): UNIVERSITY OF TORINO [IT/IT]; Department of Biomedical Sciences and Human Oncology, IRCC, SP 142, I-10060 Candiolo (IT). REGENTS OF THE UNIVERSITY OF CALIFORNIA [US/US]; 12th floor, 1111 Franklin Street, Oakland, CA 94607-5200 (US).

(84) Designated States (*regional*): ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

(72) Inventors; and

(75) Inventors/Applicants (*for US only*): ARTIGIANI, Stefania [IT/IT]; Corso Brunelleschi, 121/B, I-10100 Torino (IT). COMOGLIO, Paolo, M. [IT/IT]; Strada Valsalice, 183/8, I-10100 Torino (IT). GOODMAN, Corey, S. [US/US]; Regents of the University of California, 12th floor, 1111 Franklin Street, Oakland, CA 94607-5200 (US). TESIER-LAVIGNE, Marc [US/US]; Regents of

Published:

— Without international search report and to be republished upon receipt of that report.

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: NOVEL PLEXINS AND USES THEREOF

(57) Abstract: The invention provides methods and compositions related to novel plexins. The polypeptides may be produced recombinantly from transformed host cells and from the disclosed plexin encoding nucleic acids or purified from human cells. The invention provides isolated plexin hybridization probes and primers capable of specifically hybridizing with the disclosed plexin genes, plexin-specific binding agents such as specific antibodies, and methods of making and using the subject compositions in diagnosis, therapy and in biopharmaceutical industry. The invention also provides novel plexin neuropilin multimeric receptor complexes for semaphorins and methods of use thereof, including but not limited to, the treatment and diagnosis of neurological disease and neuroregeneration, immune modulation, and viral and oncological diseases.



WO 01/14420 A2

NOVEL PLEXINS AND USES THEREOF

BACKGROUND OF THE INVENTION

Field of the Invention

- 5 The invention relates to the identification and characterization of four novel proteins that are members of the plexin family.

Summary of the Related Art

- Plexin family members encode large transmembrane proteins, whose cysteine-rich extracellular domains share regions of homology with the Scatter Factor receptors (encoded by the Met gene family). The extracellular domains of plexins also contain ~500 amino acid Semaphorin domains (see below). The highly conserved cytoplasmic moieties of plexins (approx. 600 amino acids), however, have no homology with the Met tyrosine kinase domain, nor with any other known protein. Met-like receptors and their ligands, the Scatter Factors, mediate a complex biological program including dissociation of cell-cell contacts, motility and invasion (for a review see Tamagnone, L. and Comoglio, P.M. (1997) "Control of invasive growth by hepatocyte growth factor (HGF) and related scatter factors." *Cytokine Growth Factor Rev* 8, 129-142). During embryogenesis Scatter Factor-1 and Met promote the dissociation of cell layers in the somites and drive the migration of myogenic cells to their appropriate location (Bladt, F., Riethmacher, D., Isenmann, S., Aguzzi, A., and Birchmeier, C. (1995) "Essential role for the c-met receptor in the migration of myogenic precursor cells into the limb bud." *Nature* 376, 768-771; Maina, F., Casagrande, F., Audero, E., Simeone, A., Comoglio, P.M., Klein, R.a., and Ponzetto, C. (1996) "Uncoupling of grb2 from the met receptor in vivo reveals complex roles in muscle development." *Cell* 87, 531-542).
- 15 Met and Scatter Factor-1 are also involved in controlling neurite outgrowth and axonal guidance (Ebens, A., Brose, K., Leonardo, E.D., Hanson, M.G.J., Bladt, F., Birchmeier, C., Barres, B.A., and Tessier-Lavigne, M. (1996) "Hepatocyte growth factor/scatter factor is an axonal chemoattractant and a neurotrophic factor for spinal motor neurons." *Neuron* 17, 1157-1172; Maina, F., Hilton, M.C., Ponzetto, C., Davies, A.M., and Klein, R. (1997) "Met receptor signaling is required for sensory nerve development and HGF promotes axonal growth and survival of sensory neurons." *Genes Dev* 11, 3341-3350; Maina, F., Hilton, M.C., Andres, R., Wyatt, S., Klein, R., and Davies, A.M.
- 20
- 25
- 30

(1998) "Multiple roles for hepatocyte growth factor in sympathetic neuron development." *Neuron* 20, 835-846).

The first clue regarding a possible function for plexins came from the finding that a novel plexin, *Vespr*, interacts with the viral semaphorin A39R (Comeau, M.R., Johnson, R., DuBose, R.F., Petersen, M., Gearing, P., VandenBos, T., Park, L., Farrah, T., Buller, R.M., Cohen, J.I., Strockbine, L.D., Rauch, C., and Spriggs, M.K. (1998) "A poxvirus-encoded semaphorin induces cytokine production from monocytes and binds to a novel cellular semaphorin receptor, *VESPR*." *Immunity* 8, 473-482). Semaphorins are a large family of secreted and membrane-bound molecules that are characterized by an extracellular domain containing a ~500 amino acid Semaphorin domain (Kolodkin et al. (1993) "The semaphorin genes encode a family of transmembrane and secreted growth cone guidance molecules." *Cell* 75, 1389-1399). As noted above, plexins contain a more divergent but nevertheless conserved Semaphorin domain.

Semaphorins were originally characterized in the nervous system, where they have been implicated in repulsive axon guidance (Kolodkin et al. (1993) *supra*; Luo, Y., Raible, D., and Raper, J.A. (1993) "Collapsin: a protein in brain that induces the collapse and paralysis of neuronal growth cones." *Cell* 75, 217-227; Tessier-Lavigne, M. and Goodman, C.S. (1996) "The molecular biology of axon guidance." *Science* 274, 1123-1133). More recently, semaphorins have been furthermore implicated in cardiac and skeletal development (Behar, O., Golden, J.A., Mashimo, H., Schoen, F.J., and Fishman, M.C. (1996) "Semaphorin III is needed for normal patterning and growth of nerves, bones and heart." *Nature* 383, 525-528), in the immune response (Hall, K.T., Bousmell, L., Schultze, J.L., Boussiotis, V.A., Dorfman, D.M., Cardoso, A.A., Bensussan, A., Nadler, L.M., and Freeman, G.J. (1996) "Human CD100, a novel leukocyte semaphorin that promotes B-cell aggregation and differentiation." *Proc.Natl.Acad.Sci.U.S.A.* 93, 11780-11785), in the regulation of angiogenesis (Miao, H.Q., Soker, S., Feiner, L., Alonso, J.L., Raper, J.A., and Klagsbrun, M. (1999) "Neuropilin-1 mediates collapsin-1/Semaphorin III inhibition of endothelial cell motility. Functional competition of collapsin-1 and vascular endothelial growth factor-165" [In Process Citation]. *J Cell Biol* 146, 233-242), and in tumor growth and metastasis (Christensen, C.R., Klingelhofer, J., Tarabykina, S., Hulgaard, E.F., Kramerov, D., and Lukanidin, E. (1998) "Transcription of a novel mouse semaphorin

gene, M-semaH, correlates with the metastatic ability of mouse tumor cell lines." Cancer Res. 58, 1238-1244).

Previously identified plexins have been shown to be expressed in the developing nervous system, (i.e. Plexin-A is a receptor for class 1 semaphorins (Sema-1a and Sema-1b). Moreover, Plexin-A has been shown via genetic analysis to control motor and CNS axon guidance induced by semaphorins (Winberg, M.L., Noordermeer, J.N., Tamagnone, L., Comoglio, P.M., Spriggs, M.K., Tessier-Lavigne, M., and Goodman, C.S. (1998). Plexin A is a neuronal semaphorin receptor that controls axon guidance. Cell 95, 903-916).

Thus a need exists for discovery of other members of the plexin family of proteins.

SUMMARY OF THE INVENTION

The present invention provides four novel plexin family members.

In one aspect, the invention provides an isolated nucleic acid having at least 80% nucleic acid sequence identity to a nucleotide sequence that encodes an amino acid sequence selected from the group consisting of the amino acid sequence shown in (SEQ ID NO: 2), (SEQ ID NO: 4), (SEQ ID NO: 6) and (SEQ ID NO: 8).

In other aspects, the invention provides isolated nucleic acid having at least 80% nucleic acid sequence identity to a nucleotide sequence selected from the group consisting of the nucleotide sequence shown in (SEQ ID NO: 1), (SEQ ID NO: 3), (SEQ ID NO: 5) and (SEQ ID NO: 7).

In another aspect, the invention provides a vector comprising the nucleic acid of the above-aspects.

The invention also provides an isolated polypeptide the amino acid sequence of which comprises residues 1-18, 19-518, 451-530, 601-680, 751-830, 800-1010, or 1196-1215 of SEQ ID NO: 2; 1-23, 24-507 or 1100-1119 of SEQ ID NO: 4; or 1-42, 43-600, 541-620, 691-770, 831-910, 900-1110 or 1270-1293 of SEQ ID NO: 6; or 8-49, 154-199 or 1-199 of SEQ ID NO: 8.

In another aspect, the invention provides an isolated polypeptide having at least 80% amino acid sequence identity to an amino acid sequence selected from the group consisting of the amino acid sequence shown in (SEQ ID NO: 2), (SEQ ID NO: 4), (SEQ ID NO: 6) and (SEQ ID NO: 8)

The invention also provides a chimeric molecule comprising a polypeptide of the above aspects fused to a heterologous amino acid sequence. In one embodiment the heterologous amino acid sequence is a Fc region of an immunoglobulin.

In other aspects, the invention provides an antibody that specifically
5 binds to the polypeptides of the above aspects.

The invention also provides a method of treating, suppressing or altering a disorder involving aberrant immune regulation involving a signaling pathway between a plexin and a neuropilin in a mammal comprising the step of administering an effective amount of an agent to said mammal capable of interfering with the association between
10 the plexin and neuropilin. Contemplated agents include a chimeric molecule or an antibody of the above aspects.

DESCRIPTION OF THE DRAWINGS

Figure 1.

(A) Phylogenetic tree of human plexins. Known family members cluster in two
15 major groups: plexin A and plexin-B subfamilies. (B) Structural features of plexins, Met-like receptors and semaphorins. In the extracellular moieties, yellow boxes indicate the "sema" domains and blue boxes mark the cysteine-rich MRS motifs, some of which are stippled to indicate their atypical sequence; atypical MRS are also found in the *sema domain* of semaphorins. Sequence identity among *sema* domains ranges from 15-50%,
20 as previously described (see Winberg et al., 1998 *supra*). Potential furin-like proteolytic sites are marked by red ribbons. plexin-B1 "truncated" is the product of a splicing variant (see text). plexin-D1 and plexin-C1 (VESPR) are more distant family members, since they include atypical features in their extracellular domains. The intracellular domain of plexins (SP domain) is highly conserved through all family
25 members, and it includes two separate regions of high homology (Maestrini, E., Tamagnone, T., Longati, P., Cremona, O., Gulisano, M., Bione, S., Tamanini, F., Neel, B.G., Toniolo, D., and Comoglio, P.M. (1996) "A family of transmembrane proteins with homology to the MET-hepatocyte growth factor receptor." Proc. Natl. Acad. Sci. USA 93, 674-678) (green oval and box). Met-like receptors are disulfide-bound
30 heterodimers and include a cytoplasmic tyrosine kinase domain (red box). Mammalian semaphorins can be either secreted or cell surface proteins. Molecular weights of representative proteins are Plexin-A1 220 kDa, Plexin-B1 250 kDa, Plexin-C1 200

kDa, HGF-R/Met 145+45 kDa (heterodimer), Sema 4D 150 kDa (transmembrane), Sema7A approx. 100 kDa (membrane GPI-linked).

Figure 2.

(a) Cell surface semaphorins specifically bind human plexins. Micrographs of the binding assays done testing i) the extracellular domain of semaphorin CD100 fused to alkaline phosphatase (CD100-AP) on COS cells transfected with *plexin-B1 cDNA*; ii) control AP on plexin-B1; iii) CD100-AP on plexin-B2; iv) CD100-AP on the entire extracellular domain of plexin-B1; v) CD100-AP on isolated "plexin-B1 truncated" (including *sema* domain, 1° and 2° MRS); vi) CD100-AP on a "plexin-B1-Δsema" (including 2° and 3° MRS; vii) extracellular domain of semaphorin A39R fused to AP, on plexin-C1 (Vespr); viii) SemaK1-AP on plexin-C1. The final detection of the binding was done either using alkaline phosphatase substrates (i-vi) or by immunofluorescence (vii and viii). (B) Scatchard analysis and binding curve of CD100-AP to plexin-B1 ($K_D = 0.9 \text{ nM} \pm 0.15$).

Figure 3.

Plexins associate with neuropilins *via* specific extracellular domains. Western blots of immunoprecipitated samples from cells co-expressing neuropilins and plexins. Specific MoAbs were used, directed against the VSV-tag included in plexins or the myc-tag included in neuropilin-2 (Np2, 130 kDa). Np2 co-immunoprecipitates with plexins, such as plexin-A3 (220 kDa), the extracellular domain of plexinA1 (approx. 160 kDa), and plexin-B1 (250 kDa) but not with the unrelated cell surface receptor DCC (170 kDa). Np2 can also associate a truncated form of the extracellular moiety of plexin-B1 ("plex-B1 trunc.", approx. 110 kDa), containing the *sema domain*.

Figure 4.

Expression of mRNAs for plexins A1 (panel A, B), -A2 (panel C, D) and A3 (panel E, F) in the spinal cord (sc), dorsal root ganglia (d) and sympathetic ganglia (sg) of E13.5 mouse embryos. Expression of the mRNAs was detected by RNA in situ hybridization. Scale bar: 1 μm .

Figure 5.

Effect of a truncated plexin-A1 construct (lacking the cytoplasmic domain) on repulsive and attractive responses of *Xenopus* spinal neurons to Sema3A and netrin-1. (A-F) A control spinal neuron exposed to a gradient of Sema3A emanating from a pipette (A) is repelled away over a period of 1 hr (B). In contrast, a GFP-expressing

spinal neuron from an embryo injected with mRNA for the truncated plexin-A1 construct (C) is not affected by Sema3A (D). A similar neuron (E) shows a normal attractive response to netrin-1 (F).

(G) Cumulative distribution plot of turning angles for all the neurons studied.

- 5 Curves show the percent of neurons with turning angles less than the angle indicated on the abscissa, under different conditions (open circles, control neurons; black and blue circles, control neurons responding to Sema3A or netrin-1, respectively; red and green circles, responses of neurons expressing the truncated plexin-A1 construct to Sema3A and netrin-1, respectively. (H) Mean turning angle under all the conditions just
10 mentioned:

Figure 6.

Tyrosine phosphorylation of plexin-A3 and plexin-B1. (a) Anti-phosphotyrosine western blotting of immunoprecipitated p220^{plexin-A3} and p250^{plexin-B1} proteins. plexin-B1 is larger since it contains an extra sequence between the second and
15 the third MRS motif, in the extracellular domain (see Fig. 1). (b) The same immunoprecipitated samples underwent *in vitro* kinase assay in the presence of [γ -³²P]ATP, Mg⁺⁺ and Mn⁺⁺ ions. The SDS-PAGE was treated with alkali in conditions known to eliminate the phosphate labeling of Ser/Thr residues and specifically preserving phosphotyrosines.

20 Figure 7

Plexin-A3 overexpression mediates cell repelling cues. (a) Epithelial kidney MDCK cells transfected to overexpress plexin-A3 (or mock transfected) were cocultured with mesenchymal KJ-29 or NIH-3T3 cells. After 16-30 hours, mixed cultures of control cells (upper panels) reached confluency and stopped growing:
25 typically the epithelial cells formed islets (circled) surrounded by a fibroblasts lawn. In contrast, MDCKs overexpressing plexin-A3 (lower panels) overwhelmed the adjacent mesenchymal cells. The latter withdrew and selectively detached from the culture dish (dying cell clusters are indicated by arrowheads), and eventually epithelial cells only survived. To allow an easier detection of the mesenchymal cells, these were labeled
30 with DiI before being plated in mixed cultures. (b) Plexin-A3 expressing cells do not induce apoptotic signal on repelled fibroblasts. Mixed cultures of NIH 3T3 and control or plexin-A3 overexpressing MDCKs were tested for the presence of TUNEL-AP positive cells. Apoptotic cells were not present in clusters of repelled cells (indicated by

arrows). The right panel shows a positive control where apoptosis was induced on the same cells by UV treatment. (c) Plexin-A3 over-expressing cells form very transient contacts with fibroblasts. Time-lapse video-microscopy of control and plexin-A3 overexpressing MDCK cells grown in presence of fibroblasts. On top, snap-shot
5 images from the movie, taken every 50 minutes (real time). In the upper row is shown the persistent contact of a fibroblast (marked by an arrow) with an islet of control MDCK cells (marked by a star). In the lower row another fibroblast, instead, forms a transient contact with an islet of plexin-A3 transfected cells, which also, in turn, reshapes. At the bottom, the diagrams show the relative frequency of persistent,
10 ~~transient or very transient contacts between fibroblasts and MDCK cells.~~

DETAILED DESCRIPTION OF THE INVENTION

The reference works, patents, patent applications, and scientific literature, including accession numbers to GenBank database sequences, that are referred to herein establish the knowledge of those with skill in the art and are hereby incorporated by
15 reference in their entirety to the same extent as if each was specifically and individually indicated to be incorporated by reference. Any conflict between any reference cited herein and the specific teachings of this specification shall be resolved in favor of the later. Likewise, any conflict between an art-understood definition of a word or phrase and a definition of the word or phrase as specifically taught in this specification shall be
20 resolved in favor of the latter.

Four novel human plexins have been identified: plexin-B2, plexin-B3, plexin-D1 and Plexin A-4. Plexin-A4 is located on human chromosome 7 and is a family member of the plexin-A subfamily which also includes plexin-A1 (alternatively named plexin-1/NOV), plexin-A2 (alternatively named plexin-2/OCT) and plexin-A3 (alternatively
25 named plexin-2/SEX). Plexin-B2 and plexin-B3 are located on human chromosome 22 and chromosome X, respectively, and are family members of the plexin-B subfamily which also includes plexin-B1 (alternatively named SEP). Plexin-B3 maps very close to the plexin-A3 genomic locus on Xq28. Plexin-D1 is the first identified member of the plexin-D subfamily and is atypical of any of the other subfamilies. A fourth
30 subfamily of plexins, the plexin-C subfamily, is defined by VESPR (now plexin-C1).

The four novel plexins as described herein have in their extracellular domains regions of homology with two other protein families: (a) Scatter Factors Receptors, encoded by the *MET* oncogene family (Tamagnone and Comoglio, 1997 *supra*), and (b)

Semaphorins (Kolodkin et al. (1993) *supra* (Figure 1b). In particular, plexins and Met-like receptors contain short cysteine-rich motifs, termed "Met Related Sequences" (MRS), whose minimal consensus is: C-X(5-6)-C-X(2)-C-X(6-8)-C-X(2)-C-X(3-5)-C (Maestrini et al., 1996 *supra*); Tamagnone and Comoglio, 1997 *supra*); blue boxes in Fig. 1B). The proteins of the Met family contain a single MRS (in their receptor β chains), whereas in plexin family members there are two/three repeated MRS motifs. Furthermore, all plexin family members contain in their extracellular moiety a 500 amino acid region similar to the sema domain of semaphorins (Kolodkin et al. (1993) *supra*; Winberg et al., 1998 *supra*); yellow boxes in Fig. 1B. The MRS motif is proposed to function as protein-protein-interaction domain.

The cytoplasmic domain of plexins contains a ~600 amino acid domain which we term the SP domain ("Sex and Plexins", marked in green in Fig. 1B) that is highly conserved within the family (57-97% similarity) and in evolution (over 50% similarity between invertebrates and humans). The SP domain does not share homology with any known protein. It includes a number of potential tyrosine phosphorylation sites, but lacks the typical motifs of catalytic tyrosine kinases. Interestingly, the predicted secondary structure of the SP domain includes long conserved alpha helices, typically found in protein-protein interaction modules. Furthermore, there are several dihydrophobic amino acid motifs (such as LL or LI), known to mediate the internalization and downregulation of transmembrane receptors (Sandoval, I.V. and Bakke O. (1994). Targeting of membrane proteins to endosomes and lysosomes. Trends in Cell Biology 4, 292-297).

The present invention also demonstrates that plexins can form complexes with neuropilins, which in turn demonstrates that a receptor for semaphorins can be hetero-oligomers of plexins and neuropilins. As demonstrated by in situ mRNA expression analysis, plexins and neuropilins are in fact simultaneously expressed in several neuronal populations during embryonic development. The plexin-neuropilin complex predates ligand binding, since the association is not influenced by the presence of class 3 semaphorins. That the observed plexin-neuropilin complexes are formed in *cis* is furthermore supported by the experimental conditions used (cotransfection of isolated cells with the two constructs). An interaction in *trans* might also be envisioned (considering that plexins and semaphorins share similar *sema* domains), however by

analyzing mixed cultures of cells separately transfected with plexins and neuropilins we did not isolate associated complexes (data not shown).

We observed that the main semaphorin binding domain of neuropilins (CUB domain (Giger, R.J., Urquhart, E.R., Gillespie, S.K., Levensgood, D.V., Ginty, D.D., and Kolodkin, A.L. (1998) "Neuropilin-2 is a receptor for semaphorin IV: insight into the structural basis of receptor function and specificity." *Neuron* 21, 1079-1092; Nakamura, F., Tanaka, M., Takahashi, T., Kalb, R.G., and Strittmatter, S.M. (1998) "Neuropilin-1 extracellular domains mediate semaphorin D/III-induced growth cone collapse" [In Process Citation]. *Neuron* 21, 1093-1100; Chen et al. 1998 *supra*) is not required for the interaction with plexins, as indicated by the association of the relevant Neuropilin-2 deletion construct with plexin-B1 (not shown). A ternary complex, where neuropilins use two distinct protein modules to form a bridge between the sema domain of semaphorins and the sema domain of plexins is thus contemplated. It is further contemplated that plexins are the functional partners of neuropilins, required for transducing signals mediated by semaphorins, preferably class 3 semaphorins. For example, in flies, which lack both neuropilins and class 3 semaphorins, D Plex A appears sufficient as a functional receptor for Sema 1a, a transmembrane class 1 semaphorin (Winberg et al., 1998 *supra*). Further support that plexins are functional co-receptors for secreted semaphorins is demonstrated in an experiment that shows that a truncated plexin-A1 construct expressed in *Xenopus* spinal neurons abolishes repulsive responses to Sema3A without markedly affecting attractive responses to netrin-1. These results are consistent with the involvement of plexins.

The intracellular signals transduced by plexins are still largely obscure. The cytoplasmic domain of plexins is large and highly conserved within and across species and contains stretches of alpha helices, which are putative protein-protein interaction domains, and could thus mediate the association with cytosolic partners. We demonstrate herein that the cytoplasmic domain of plexins can be tyrosine phosphorylated, suggesting that, like other receptors devoid of intrinsic catalytic activity, plexins may signal by associating a tyrosine kinase (Stahl, N. and Yancopoulos, G.D. (1993). The alphas, betas, and kinases of cytokine receptor complexes. *Cell* 74, 587-590; Glass, D.J., Bowen, D.C., Stitt, T.N., Radziejewski, C., Bruno, J., Ryan, T.E., Gies, D.R., Shah, S., Mattsson, K., Burden, S.J., DiStefano, P.S.,

Valenzuela, D.M., DeChiara, T.M., and Yancopoulos, G.D. (1996). Agrin acts via a MuSK receptor complex. *Cell* 85, 513-523).

In addition, we show herein that expression of plexins, particularly plexin-A3, mediates cell-repelling cues. By time-lapse video-microscopy we observed a true
5 repelling effect on fibroblasts. Intriguingly, we observed that -upon interaction with fibroblasts- also the islets of plexin-A3 MDCKs at times reshaped. This may be explained by the existence of intra-epithelial repelling cues, balanced by the attractive forces exerted by epithelial cell junctions.

Moreover we have demonstrated that in the nervous system (i.e. *Drosophila*), that
10 defasciculating motor axons co-express both Plexin A and one of its interacting partners, the transmembrane semaphorin Sema-1a (Winberg et al., 1998 *supra*). This demonstrates that plexins act *in vivo* either as receptors or ligands for cell surface semaphorins, which in turn can transduce intracellular signals, as reported for ephrins (Holland et al., 1996 *supra*). Semaphorins, therefore, besides being pivotal in axon
15 guidance, have a general role in morphogenesis and tissue remodeling by mediating cell-repelling cues via their interactions with plexins.

Accordingly, in a first aspect, the invention provides an isolated nucleic acid molecule encoding a novel human plexin polypeptide. By "plexin polypeptide" is meant a member of the plexin family comprising an amino acid sequence that shares at least
20 60% amino acid sequence homology with SEQ ID NOS: 2 (plexin B-2), 4 (plexin B-3), 6 (plexin D-1) or 8 (plexin A-4), preferably, at least 65% sequence homology, more preferably, at least 70% sequence homology, more preferably, at least 75% sequence homology, more preferably, at least 80% sequence homology, still more preferably at least 85% sequence homology, even more preferably, at least 90% sequence
25 homology, and most preferably at least 95% sequence homology with SEQ ID NOS: 2, 4, 6 or 8. Plexin polypeptides of the invention are useful for modulating cell growth (i.e. nerve) and immune regulation.

As used herein, by "modulating" is meant increasing or decreasing cell growth. By "cell growth" is meant any change in cell number or size, including, without
30 limitation, increase or decrease in cell number, increase or decrease in rate of cell division, increase or decrease in rate of cell death, and/or increase or decrease in cell size. Standard methods for measuring cell growth include standard apoptosis assays (e.g., TUNEL assays, DNA fragmentation, trypan blue exclusion) and cell proliferation assays

(e.g., ³H-thymidine incorporation). It will be appreciated that the degree of modulation of cell growth provided by a plexin polypeptide in a given assay will vary, but one of skill in the art can readily determine the statistically significant change in cell growth of a cell exposed to a plexin polypeptide.

5 By "immune regulation" is meant increasing or decreasing the biological functions of immune cells (i.e., cells involved in an immune response). Immune cells include, without limitation, lymphocytes (T and B), NK cells, dendritic cells, myeloid cells (e.g., macrophages and neutrophils), and other hematopoietic cells involved in an immune response.

10 By "nucleic acid molecule" or "nucleic acid" as used herein, is meant any deoxyribonucleic acid (DNA) or ribonucleic acid (RNA), including, without limitation, complementary DNA (cDNA), genomic DNA, RNA, heteronuclear RNA (hnRNA), messenger RNA (mRNA), DNA/RNA hybrids, or synthetic nucleic acids (e.g., an oligonucleotide) comprising ribonucleic and/or deoxyribonucleic acids or synthetic
15 variants thereof. The nucleic acid of the invention includes, without limitation, an oligonucleotide or a polynucleotide. The nucleic acid can be single stranded, or partially or completely double stranded (duplex). Duplex nucleic acids can be homoduplex or heteroduplex.

By "polypeptide" is meant any molecule comprising two or more amino acids
20 joined together with a peptide bond, regardless of length or post-translational modifications (e.g., without limitation, glycosylation, lipidation, acetylation, or phosphorylation). Useful plexin polypeptides of the invention include, without limitation, the full length plexin polypeptides having the amino acid sequence of SEQ ID NOS: 2, 4, 6, 8 or 10; an extracellular domain of the polypeptide having the amino
25 acid sequence 1 to 1199 of SEQ ID NO: 2; 1 to 1099 of SEQ ID NO: 4; 1 to 1270 of SEQ ID NO: 6 or 1 to 199 of SEQ ID NO: 8, with its associated signal peptide; or an extracellular domain of the polypeptide having the amino acid sequence 19 to 1199 of SEQ ID NO: 2; 24 to 1099 of SEQ ID NO: 4; or 43 to 1270 of SEQ ID NO: 6, lacking its associated signal peptide; an intracellular domain of the polypeptide having the
30 amino acid sequence of SEQ ID NOS: 2, 4, 6 or 8; and polypeptides, the amino acid sequence of which comprises about residues 1-18 (putative signal sequence), 19-518 (sema domain), 451-530 (1° MRS), 601-680 (2° MRS), 751-830 (3° MRS), 800-1010 (G-P repeats) or 1196-1215 (putative transmembrane domain) of SEQ ID

NO: 2; about residues 1-23 (putative signal sequence), 24-507 518 (sema domain) or 1100-1119 (putative transmembrane domain) of SEQ ID NO: 4; or about residues 1-42 (putative signal sequence), 43-600 (sema domain), 541-620 (1° MRS), 691-770 (2° MRS), 831-910 (3° MRS), 900-1110 (G-P repeats) or 1270-
5 1293 (putative transmembrane domain) of SEQ ID NO: 6; or about residue 8-49 (1° MRS) or 154-199 (2° MRS) of SEQ ID NO: 8.

By "isolated" is meant a compound (*e.g.*, a nucleic acid molecule or a protein) that has been separated from components (*e.g.*, nucleic acid molecules, proteins, lipids, and/or carbohydrates) which naturally accompany it. Water, buffers, and other small
10 molecules (*e.g.*, ~~molecules having a molecular weight of less than about 1000 daltons~~) may accompany an isolated compound of the invention. Preferably, an isolated compound is at least 70%, by weight, free from components which naturally accompany it. More preferably, an isolated is at least 75%, by weight, free from components which naturally accompany it; still more preferably, at least 80%, by
15 weight, free; even more preferably, at least 85%, by weight, free; and even more preferably, at least 90%, by weight, free from components which naturally accompany it. Most preferably, a substantially purified compound is at least 95%, by weight, free from components which naturally accompany it.

Where the isolated compound is a nucleic acid molecule, the isolated nucleic acid
20 molecule is separated from other nucleic acids (*e.g.*, genes or transcripts) or proteins which, in the naturally-occurring genome of the organism from which the nucleic acid molecule was derived, flanked the nucleic acid molecule. Isolated nucleic acid molecules therefore include, without limitation, a recombinant nucleic acid molecule incorporated into a plasmid or other vector (*e.g.*, a replication-defective virus); a
25 recombinant nucleic acid molecule incorporated into the genome of a prokaryotic or eukaryotic organism; or a nucleic acid molecule which exists as a separate molecule independent of other nucleic acids (*e.g.*, a PCR product, a chemically synthesized nucleic acid molecule, or a nucleic acid molecule produced by restriction endonuclease digestion). Purification of a nucleic acid molecule can be accomplished and measured by
30 any standard method including, without limitation, sequence analysis, chemical synthesis, PCR, CsCl gradient, phenol:chloroform extraction, ethanol precipitation, Southern or Northern blotting analysis followed by band extraction and purification, and

high performance liquid chromatography (HPLC; see, *e.g.*, Fisher (1980) Laboratory Techniques in Biochemistry and Molecular Biology, Work and Burdon (eds.), Elsevier).

Thus, in one non-limiting example, to obtain an isolated nucleic acid molecule encoding a plexin polypeptide, a nucleic acid molecule is chemically synthesized on a standard oligonucleotide synthesis machine. The resulting single stranded molecule is then subjected to second strand synthesis to form a duplex DNA molecule, which is then ligated into a plasmid capable of replication in a prokaryotic or eukaryotic cell. The nucleic acid molecule is then replicated in the cell, purified (*e.g.*, by CsCl gradient), and subjected to sequence analysis.

~~In certain embodiments of the first aspect of the invention, the nucleic acid molecule has a nucleic acid sequence comprising SEQ ID NOS: 1, 3, 5, 7 or 9. Preferably, the nucleic acid molecule of the invention has not more than 500 nucleotides flanking each of the 5' and 3' ends of SEQ ID NOS: 1, 3, 5, 7 or 7. In certain embodiments, the plexin polypeptide has an amino acid sequence that comprises SEQ ID NOS: 2, 4, 6, 8 or 10. Preferably, the plexin polypeptide of the invention has not more than 50 amino acid residues flanking each of the N-terminal and C-terminal ends of SEQ ID NOS: 2, 4, 6, 8 or 10.~~

In certain embodiments of the first aspect of the invention, the nucleic acid molecule hybridizes under stringent conditions (as defined herein) to SEQ ID NOS: 1, 3, 5, 7 or 9.

The invention also includes nucleic acid molecules that hybridize under stringent hybridization conditions (as defined herein) to all or a portion of the nucleotide sequence represented by SEQ ID NOS: 1, 3, 5, 7 or 9 or its complement. The hybridizing portion of the hybridizing nucleic acid is at least 80%, *e.g.*, at least 95%, or at least 98%, homologous to the sequence of a portion or all of a nucleic acid encoding a polypeptide having the amino acid sequence of SEQ ID NOS: 2, 4, 6, 8 or 10, or its complement. Hybridizing nucleic acids of the type described herein can be used, for example, as a cloning probe, a primer (*e.g.*, a PCR primer) or a diagnostic probe.

Hybridization of the oligonucleotide probe to a nucleic acid sample typically is performed under stringent conditions. Nucleic acid duplex or hybrid stability is expressed as the melting temperature or T_m , which is the temperature at which a probe dissociates from a target DNA. This melting temperature is used to define the required stringency conditions. If sequences are to be identified that are related and substantially

identical, rather than identical, then it is useful to first establish the lowest temperature at which only homologous hybridization occurs with a particular concentration of salt (e.g., SSC or SSPE). Then, assuming that 1% mismatch results in a 1°C decrease in the T_m , the temperature of the final wash in the hybridization reaction is reduced accordingly (for example, if the sequences have > 95% identity with the probe are sought, the final wash temperature is decreased 5°C). In practice, the change in the T_m can be between 0.5 C and 1.5 C per 1% mismatch. "Stringent conditions" involve hybridization at 68°C in 5x SSC/5x Denhardt's solution/1.0% SDS, and washing in 0.2xSSC/0.1% SDS at room temperature. "Moderately stringent conditions" include washing in 3xSSC at 42°C. The parameters of salt concentration and temperature can be varied to achieve the optimal level of identity between the probe and the target nucleic acid. Additional guidance regarding such conditions is readily available in the art, for example, by Sambrook *et al.*, *supra*; and Ausubel *et al.*, *supra*.

Nucleic acid sequence homology (as well as amino acid sequence homology) can be measured according to standard methods. Unless otherwise specified, as used herein used herein, "percent homology" of two amino acid sequences or of two nucleic acids is determined using the algorithm of Karlin and Altshul (*Proc. Natl. Acad. Sci. USA* 87: 2264-2268, 1990), modified as in Karlin and Altschul (*Proc. Natl. Acad. Sci. USA* 90: 5873-5877, 1993). Such an algorithm is incorporated into the NBLAST and XBLAST programs of Altschul et al. (*J. Mol. Biol.* 215: 403-410, 1990). BLAST nucleotide searches are performed with the NBLAST program, e (score) = 100, word length = 12, to obtain nucleotide sequences homologous to a nucleic acid molecule of the invention. BLAST protein searches are performed with the XBLAST program, e (score) = 50, word length = 3, to obtain amino acid sequences homologous to a reference polypeptide (e.g., SEQ ID NO: 2). To obtain gapped alignments for comparison purposes, Gapped BLAST is utilized as described in Altschul et al. (*Nucleic Acids Res.* 25: 3389-3402, 1997). When utilizing BLAST and Gapped BLAST programs, the default parameters of the respective programs (e.g., XBLAST and NBLAST) are used, namely e=10; w=11 for nucleic acid; w=3 for amino acid (the Blossum 62 scoring matrix); low complexity sequence filtering. The default settings of BLAST emphasize regions of local alignment to detect relationships among sequences which share only isolated regions of similarity (Altschul et al., *J. Mol. Biol.* 215: 403-410 (1990). See <http://www.ncbi.nlm.nih.gov>.

Thus, in a non-limiting example to obtain an isolated nucleic acid molecule encoding a plexin polypeptide, a nucleic acid molecule having the sequence of SEQ ID NOS: 1, 3, 5 or 7 is used to probe a cDNA library under stringent conditions according to standard techniques (see., e.g., Ausubel *et al.*, *supra*). Upon identification of a positive
5 clone (*i.e.*, a clone that hybridizes to SEQ ID NOS: 1, 3, 5 or 7 under stringent conditions), that clone is expanded and subjected to sequence analysis. A nucleic acid molecule having a nucleic acid sequence that is at least 70% identical, preferably at least 75% identical, more preferably, at least 80% identical, still more preferably at least 85% identical, even more preferably, at least 90% identical, and most preferably at least 95%
10 identical (as measured by the basic BLAST program of NCBI on default settings) to SEQ ID NOS: 1, 3, 5 or 7 is a nucleic acid molecule of the invention.

In a second aspect, the invention provides four novel isolated plexin polypeptides.

"Isolated" is as defined for the first aspect of the invention. Where the isolated compound is a polypeptide, the isolated polypeptide is separated from organic molecules,
15 such as nucleic acid molecules, polypeptides, and/or carbohydrates, which, in the naturally-occurring organism from which the polypeptide was derived, accompany the polypeptide. Isolated polypeptides therefore also include a recombinant polypeptide (*e.g.*, a human polypeptide expressed in an insect cell), or a chemically synthesized polypeptide. Purification of a polypeptide can be accomplished and measured by any
20 standard method including, without limitation, chemical synthesis, recombinant polypeptide expression in prokaryotic or eukaryotic cells, affinity chromatography, Western blotting analysis, SDS-PAGE analysis, and/or HPLC.

In accordance with this aspect, the invention provides all derivatives, mutants, truncations, and/or splice variants of the four novel plexin polypeptides, so long as these
25 derivatives, mutants, truncations, and/or splice variants share at least 60% amino acid sequence homology with SEQ ID NOS: 2, 4, 6 or 8, preferably, at least 65% sequence homology, more preferably, at least 70% sequence homology, more preferably, at least at least 75% sequence homology, more preferably, at least 80% sequence homology, still more preferably at least 85% sequence homology, even more preferably, at least 90%
30 sequence homology, and most preferably at least 95% sequence homology with SEQ ID NOS: 2, 4, 6 or 8 as determined using the basic BLAST program of the National Center for Biotechnology (NCBI; National Library of Medicine, Bethesda, MD), using the

default settings defined therein using the sequence of the four novel plexin derivative, mutant, truncation and/or splice variance as the probe.

Preferred plexin polypeptide derivatives include polypeptides whose sequences differ from the sequence given in SEQ ID NOS: 2, 4, 6 or 8, by one or more conservative amino acid substitutions, or by one or more non-conservative amino acid substitutions, deletions or insertions which do not abolish the biological activity of the plexins. Conservative amino acid substitutions typically include the substitution of one amino acid for another with similar biochemical characteristics, such as polarity, size, and/or charge. Non-limiting examples of conservative substitutions are substitutions within the following groups: valine, glycine, alanine, valine, isoleucine, leucine; aspartic acid, glutamic acid, asparagine, glutamine; serine, threonine; lysine, arginine, phenylalanine, and tyrosine.

Useful methods for mutagenesis to generate plexin mutants are known in the art (see, e.g., Sambrook *et al.*, *supra*; Ausubel *et al.*, *supra*). Preferred methods for mutagenesis are described in PCT Publication WO99/12965 and include PCR mutagenesis and saturation mutagenesis. A library of random amino acid sequence variants can also be generated by the synthesis of a set of degenerate oligonucleotide sequences.

In certain embodiments of the second aspect of the invention, the plexin polypeptide has a sequence comprising the sequence of SEQ ID NOS: 2, 4, 6 or 8. In one non-limiting example, in accordance with the invention, an isolated plexin polypeptide comprising the sequence of SEQ ID NOS: 2, 4, 6 or 8 can chemically synthesized according to standard techniques (e.g., at a commercial peptide generating facility).

For example, a putative plexin polypeptide is purified and subjected to N-terminal sequencing to determine its amino acid sequence. The amino acid sequence of the polypeptide is then compared to SEQ ID NOS: 2, 4, 6, 8 or 10 (as measured by the basic BLAST program of NCBI on default settings). A polypeptide that shares at least 60% homology with SEQ ID NOS: 2, 4, 6, 8 or 10 is a plexin polypeptide of the invention.

In another example, purification of a plexin polypeptide is facilitated by the addition of a tag to the polypeptide that enables purification of the tagged polypeptide. Non-limiting examples of a tag include a hemagglutinin (HA) tag, a his tag, a GST tag, a

FLAG-tag, and a myc tag. Thus, a nucleic acid molecule of the first aspect is engineered using standard molecular biology techniques to incorporate the nucleic acid sequence encoding the tag. The engineered nucleic acid molecule is then introduced and positioned for expression in an appropriate cell to produce the recombinant tagged polypeptide, which can then be readily purified by binding of the tag to its substrate. For example, the his tag binds to Ni-NTA agarose. Likewise, a GST (glutathione S-transferase) tag binds to glutathione agarose beads. Both his tag and GST tag expression and purification kits are commercially available from PharMingen (San Diego, CA). Likewise, myc-tagged plexin polypeptide are produced by cells introduced with a nucleic acid molecule encoding the tagged protein and positioned for expression in the cell.

It will be appreciated that particularly useful polypeptides of this aspect of the invention are secreted by the cell in which they are produced, thus facilitating purification of the polypeptide from the culture media in which the cells have been maintained, without requiring lysis of the cell.

In a third aspect, the invention provides a cell engineered to comprise a nucleic acid molecule encoding one of the four plexin polypeptides. By "engineered" is meant that the cell of the invention has been modified by standard molecular biology techniques. Where the cell is "engineered to comprise a nucleic acid molecule," standard molecular biology techniques have been employed to introduce the indicated nucleic acid molecule into the cell, either by transformation or transfection of the cell with a plasmid, or by infection or transduction of the cell with a recombinant virus.

The nucleic acid molecule of the first aspect of the invention is preferably subcloned into a plasmid or vector (for example, but not limited to, a vector used to generate a recombinant virus), wherein the nucleic acid molecule is positioned for expression in the plasmid or vector. The plasmid or vector is then introduced into a cell by standard techniques to produce an engineered cell in accordance with the third aspect of the invention.

In certain embodiments of the third aspect, the cell is a prokaryotic cell (*e.g.*, a bacterium). For example, *E. coli* cells (*e.g.*, DH5 α) are transformed (using, *e.g.*, electroporation) with a bacterial plasmid (*i.e.*, a plasmid containing an *E. coli* origin of replication) containing a nucleic acid molecule of the first aspect of the invention. The transformed bacteria are selected using, for example, an antibiotic-resistance encoding nucleic acid molecule (*e.g.*, ampicillin resistance) on the plasmid such that the antibiotic

resistance protein is expressed by the transformed bacteria. The transformed bacteria are then propagated, and can be cryopreserved and stored frozen in glycerol.

Those of skill in the art will appreciate that in accordance with the third aspect of the invention, a nucleic acid molecule encoding one of the four plexin polypeptides may be introduced into a large variety of cells. For example, a nucleic acid molecule encoding one of the four plexin polypeptides can be introduced into prokaryotic cells (e.g., bacteria), and any eukaryotic cell into which an exogenous nucleic acid molecule may be introduced. Thus, in certain embodiments of the third aspect of the invention, the cell is a eukaryotic cell. Eukaryotic cells according to this aspect of the invention that comprise a nucleic acid molecule encoding one of the four plexin polypeptides include, without limitation, yeast cells, plant cells, insect cells, and mammalian cells. Within the category of mammalian cells are cells from any mammalian species (including, without limitation, mouse, hamster, monkey, human), of any lineage (including, without limitation, lymphocyte, fibroblast, stem cell), and may be an immortalized cell, or a non-immortalized cell. Cells, as well as plasmids and/or vectors (e.g., vectors that can be packaged to form infectious virus particles), are commercially available, for example, from the American Type Culture Collection ("ATCC"; Manassas, VA).

In certain embodiments of the third aspect of the invention, the nucleic acid molecule is positioned for expression in the cell. By "positioned for expression" is meant that the nucleic acid molecule is operably linked to at least one regulatory sequence which directs the transcription and translation of the nucleic acid molecule in a cell, such that the cell engineered to comprise the nucleic acid molecule produces (i.e., expresses) the protein encoded by the nucleic acid molecule. By "operably linked" is meant that the nucleic acid molecule and the regulatory sequence are connected in a such a way as to permit expression of the nucleic acid molecule when the nucleic acid molecule is present in a cell. Regulatory sequences include, without limitation, promoters, enhancers, IRES sequences, and polyadenylation signals. Since plexin polypeptides are involved in immune regulation and the modulation of cell growth, it may be desirable to operably link a nucleic acid molecule encoding one of the four plexin polypeptides to an inducible promoter.

The four plexin polypeptides that are encoded by the nucleic acid molecules do not necessarily include the transmembrane domain of the four plexin polypeptides, and so may be produced by the cell as an intracellular polypeptide or a soluble secreted

polypeptide. For example, if the polypeptide fragment is secreted by the cell, it can be purified from the conditioned growth media of the transfected cells, without having to lyse the cells. Likewise, although a soluble intracellular polypeptide fragment is purified from only lysed cells, the fragment, being soluble, does not have to be extracted from the cell membrane; thus, different lysis conditions may be used to obtain purified soluble intracellular polypeptide fragment as compared to the lysis conditions required to obtain purified full length plexin polypeptides (which has a transmembrane domain).

Protein expression systems have been established for a variety of cells and are known to those of skill in the art. Cells are also commercially available from the ATCC, and a variety of protein expression system kits are commercially available from, for example, Invitrogen Corp. (Carlsbad, CA), Clontech Laboratories (Palo Alto, CA), PharMingen (San Diego, CA), Promega Corp. (Madison, WI), and Stratagene (La Jolla, CA).

For example, a nucleic acid molecule encoding one of the four plexin polypeptides is operably linked to bacterial regulatory sequences (e.g., T7 late promoter or bacteriophage regulatory sequences), and the resulting nucleic acid molecule is used to transform bacterial cells, where the transformed bacterial cells produce one of the four plexin polypeptides. In another example, a nucleic acid molecule encoding one of the four plexin polypeptides is operably linked to baculovirus regulatory sequences in a baculovirus vector. Recombinant baculovirus are then generated and used to transduce insect cells (using, for example, the expression kit commercially available from Clontech Laboratories). The transduced insect cells comprise a nucleic acid molecule encoding one of the four plexin polypeptides positioned for expression in the insect cell, and thus produce one of the four plexin polypeptides.

Mammalian cells are widely used as protein expression systems. For example, a mammalian cell may be transduced with a recombinant retrovirus or adenovirus comprising a nucleic acid molecule encoding one of the four plexin polypeptides operably linked to regulatory sequences that are either endogenous to the particular virus or exogenous to the virus (e.g., a CMV promoter in a retroviral vector). The transduced mammalian cell is then propagated *in vitro* in tissue culture, *in vivo* (e.g., in an immunocompromised animal), and/or cryopreserved and stored frozen in DMSO.

In another example, mammalian cells are transfected with an expression plasmid comprising a nucleic acid molecule encoding one of the four plexin polypeptides

operably linked to one or more regulatory sequences on the plasmid. By "expression plasmid" is meant a plasmid in which an inserted nucleic acid molecule of interest (*e.g.*, encoding one of the four plexin polypeptides, a plexin chimeric molecule, or tagged plexin polypeptide) is operably linked to at least one regulatory sequence such that when
5 the expression plasmid containing the inserted nucleic acid molecule of interest is introduced (*e.g.*, by transfection) into a cell, the inserted nucleic acid molecule is positioned for expression in that cell. The nucleic acid molecule in the expression plasmid, upon being introduced into the cell, is thus positioned for expression in that cell, and enables the cell to produce one of the four plexin polypeptides encoded by the
10 nucleic acid molecule.

In one non-limiting example, a nucleic acid molecule according to the first aspect of the invention is inserted into a standard mammalian expression plasmid (*e.g.*, pcDNA3.1 commercially available from Invitrogen Corp., Carlsbad, California), such that the inserted nucleic acid molecule encoding one of the four plexin polypeptides is
15 operably linked to the regulatory sequences in the mammalian expression plasmid. Mammalian cells are then transfected with this expression plasmid (using, *e.g.*, CaPO_4 or DEAE-dextran). Where the expression plasmid contains an antibiotic-resistance encoding nucleic acid molecule (*e.g.*, neomycin resistance on the pcDNA3.1 plasmid) such that the antibiotic resistance protein is expressed by the transfected cells, transfected
20 cells may be selected for the ability to grow in the presence of the antibiotic. The transfected cells may then be propagated and cryopreserved and stored in frozen in DMSO.

In a fourth aspect, the invention provides an isolated nucleic acid molecule encoding a chimeric molecule comprising at least two segments, wherein one of the
25 segments comprises one of the four plexin polypeptides. By "chimeric molecule" is meant a protein that comprises at least two segments of polypeptide joined together by any means, including, without limitation, a covalent bond (*e.g.*, peptide bond), a non-covalent bond (*e.g.*, ionic bond or hydrogen bond) or by a chemical crosslinker. It should be noted that one of the four plexin polypeptides that has been tagged is within the
30 definition of a chimeric molecule.

In certain embodiments of the fourth aspect of the invention, the nucleic acid molecule encoding the segment of a chimeric molecule comprising one of the four plexin

polypeptides hybridizes under stringent conditions to SEQ ID NO: 1, 3, 5 or 7.

"Stringent conditions" are as described above for the first aspect of the invention.

Standard molecular biology techniques may be employed to generate nucleic acid molecules encoding chimeric molecules according to the fourth aspect of the invention.

- 5 For example, a nucleic acid molecule encoding the extracellular domain of one of the four plexin polypeptides may be joined, in frame, to a nucleic acid molecule encoding the constant region of an immunoglobulin molecule (see, *e.g.*, Chamow S.M., Antibody Fusion Proteins, John Wiley & Sons, New York, 1999). By "in frame" is meant that a first nucleic acid molecule is ligated to a second nucleic acid molecule such that the each
10 of the amino acid sequences of the polypeptides encoded by each of the first and the second nucleic acid molecules is not frame-shifted.

- In one non-limiting example, a chimeric molecule comprising the extracellular domain of one of the four plexin polypeptides including the amino acid sequence of SEQ ID NOS: 2, 4, 6 or 8 is generated. In this example, a nucleic acid molecule encodes
15 amino acid residue number 1(19) through about amino acid residue number 1199 of SEQ ID NO: 2; amino acid residue number 1(24) through about amino acid residue number 1099 of SEQ ID NO: 4; amino acid residue number 1(43) through about amino acid residue number 1270 of SEQ ID NO: 6 and amino acid residue number 1 through about amino acid residue number 199 of SEQ ID NO: 8 with its associated signal peptide
20 (parenthesis depicts about the beginning of the amino acid sequence of the extracellular domain lacking its signal peptide). This nucleic acid molecule is fused in frame with a nucleic acid molecule encoding the constant region of an immunoglobulin, such that the chimeric molecule encoded by the resulting nucleic acid molecule generally has the following structure:

25

N-terminus	extracellular domain of SEQ ID NO: 2 with or lacking its signal peptide	amino acids from the constant region of an Ig molecule	C-terminus
N-terminus	extracellular domain of SEQ ID NO: 4 with or lacking its signal peptide	amino acids from the constant region of an Ig molecule	C-terminus
N-terminus	extracellular domain of SEQ ID NO: 6 with or lacking its signal peptide	amino acids from the constant region of an Ig molecule	C-terminus
N-terminus	extracellular domain of SEQ ID NO: 8 with or lacking its signal	amino acids from the constant region of an Ig molecule	C-terminus

	peptide		
--	---------	--	--

The heavy chain class (*e.g.*, IgG, IgA, IgM, IgD, or IgE) can be varied in this chimeric molecule depending upon which constant region is used. Nucleic acid molecules encoding the constant region of various immunoglobulin (Ig) heavy chains are known
 5 (see, *e.g.*, Zettlmeissl et al., *DNA Cell Biol.* **9**(5):347-53, 1990) Indeed, expression plasmids are available, into which the nucleic acid molecule of interest (*i.e.*, a nucleic acid molecule encoding an extracellular domain of the polypeptide of SEQ ID NO: 2; SEQ ID NO: 4; SEQ ID NO: 6; or SEQ ID NO: 8) can be inserted, and the resulting
 plasmid introduced into a cell to produce one of the four extracellular plexin-Ig chimeric
 10 molecule s (see, *e.g.*, Zettlmeissl et al., *supra*; Miller et al., *J. Exp. Med.* **178** (1): 211-222, 1993).

Any variety of chimeric molecule carrying the extracellular domains of one of the four plexin polypeptide may be generated. For example, the extracellular domain of one of the four plexin polypeptides can be myc-tagged, his-tagged, or FLAG tagged
 15 according to standard molecular biology techniques.

Such extracellular proteins are particularly useful for identifying ligands to which the extracellular domain of one of the four plexin polypeptides bind. For example, extracellular plexin-D1-Ig chimera can be immobilized on a protein A-sepharose column. Molecules suspected of binding the extracellular domain of plexin-D1 are added to the
 20 column, to which the molecule that binds to the extracellular domain of plexin-D1 adhere, and the non-binding molecules flow through the column. The extracellular plexin-D1-binding molecules are readily eluted, for example, by changing the pH or ion concentration of the elution buffer.

Extracellular plexin proteins are also used to identify cells expressing the ligand
 25 of plexin extracellular domain on their cell surface (and thereby also identify the ligand itself). For example, cells are incubated with a FLAG-tagged plexin extracellular domain chimeric molecule. A FLAG-tagged plexin extracellular domain chimeric molecule is generated. An anti-FLAG antibody that is detectably labeled is then added to the cells. By "detectably labeled" is meant any means for marking and identifying the presence of a
 30 molecule. Detectable labels include, without limitation, radioactive labels (*e.g.*, ³²P or ³⁵S) and fluorophore labels (*e.g.*, FITC, phycoerythrin, or rhodamine). For example, FITC-labeled anti-FLAG antibodies are commercially available from Babco, Richmond,

CA. The "stained" cells (*i.e.*, cells incubated first with the FLAG-tagged plexin extracellular domain chimeric molecule and then with the FITC-labeled anti-FLAG antibody), are then subjected to flow cytometry analysis to select those cells that are labeled with FITC, and so express a molecule that binds to the extracellular domain of one of the four plexin polypeptides. The FITC labeled cells are then further manipulated (e.g., characterized to determine which cells express the plexin polypeptide ligand).

The ligand of the plexin extracellular domain is itself identified, for example, by lysing the cells, adding the lysate to one of the four plexin extracellular domain-Ig chimeric molecule columns described above, and purifying the ligand. The ligand is then sequenced by N-terminal sequencing.

In another non-limiting example, the intracellular domain of one of the four plexin polypeptides is used as "bait" in a yeast two-hybrid system to identify ligands that interact with the intracellular domain of one of the four plexins described herein. For general description of the two-hybrid system, see U.S. Patent Nos. 5,283,173; 5,468,614; and 5,695,941. In this example, a nucleic acid molecule encoding from about amino acid residue number 143 through at least amino acid residue number 214 of SEQ ID NO: 2 is inserted into a standard DNA binding domain expression plasmid (e.g., the GAL4 DNA binding domain plasmid in the Interactor kit commercially available from PharMingen (San Diego, CA). (It will be understood that the nucleic molecule may encode amino acid residue number 138-148 through at least amino acid residue number 214 of SEQ ID NO: 2.) A variety of cDNA libraries in transcriptional activation domain vectors are available (e.g., from Clontech, Palo Alto CA). The cDNA libraries are screened employing standard methods (see, e.g., the methods employed in U.S. Patent No. 5,780,262) to identify cDNA clones encoding a ligand that binds to the intracellular domain of one of the four plexin polypeptides. One preferable cDNA library screened in this example is a cDNA library generated from an immune cell (e.g., a lymphocyte or NK cell).

In a fifth aspect, the invention provides a purified chimeric molecule comprising one of the four plexin polypeptides. Methods for purifying proteins are as described for the second aspect of the invention.

In a sixth aspect, the invention provides a cell engineered to comprise a nucleic acid molecule encoding a chimeric molecule comprising at least two segments, wherein one of the segments comprises one of the four plexin polypeptides. As described for the

third aspect of the invention, a nucleic acid encoding a chimeric molecule comprising one of the four plexin polypeptides may be introduced into any variety of cells. In certain embodiments, the cell is a prokaryotic cell or a eukaryotic cell. In certain embodiments, the eukaryotic cell is a yeast cell or a mammalian cell (*e.g.*, a human cell).

5 In a seventh aspect, the invention provides an isolated binding agent that specifically binds one of the four plexin polypeptides, or specifically binds a chimeric molecule comprising a segment comprising one of the four plexin polypeptides. In certain embodiments, the plexin protein has an amino acid sequence comprising SEQ ID NOS:2, 4, 6 or 8.

10 By "specifically binds" is meant a binding agent (*e.g.*, an antibody) that binds to its specific target (*e.g.*, one of the four plexin polypeptides) with greater affinity than it binds to other molecules. Preferably, where the binding agent is an antibody, the antibody preferably specifically binds to its specific target with a dissociation constant (K_D) of at least 10^{-5} M, more preferably, 10^{-6} M, even more preferably 10^{-7} M, and most preferably, the binding agent specifically binds to its specific target with a K_D of at least 10^{-8} M.

Preferably, the binding agent of this aspect of the invention is an antibody, such as a monoclonal antibody or a polyclonal antibody, or a fragment of an antibody that specifically binds one of the four plexin polypeptides. Standard methods may be
20 employed to generate both monoclonal and polyclonal antibodies that specifically bind to one of the four plexin polypeptides of the invention. See, *e.g.*, Ausubel et al., *supra*; Coligan, J.E. et al., Current Protocols in Immunology, John Wiley & Sons, New York (1991); and Delves, P.J., Antibody Production: Essential Techniques, John Wiley & Sons, New York (1997). Briefly, the plexin polypeptides of the present invention,
25 purified according to the methods described for the second aspect of the invention, are used to immunize rabbits (*e.g.*, for polyclonal antibodies) or mice (*e.g.*, for monoclonal antibodies) to generate antibody-mediated immunity to the four plexin polypeptides used to immunize the animal. For monoclonal antibodies, antibodies can be screened by, *e.g.*, ELISA, to identify those antibodies that show the highest affinity for the immunizing
30 plexin protein of polypeptide fragment. The cloned cell producing the high affinity monoclonal antibody can then be propagated *in vitro* (where the antibody is purified from the culture supernatant) or *in vivo* (where the antibody is purified from ascites fluid), and

can also be cryopreserved and stored frozen at, *e.g.*, -70°C in DMSO, to provide a potentially limitless supply of monoclonal antibody.

In addition to intact monoclonal and polyclonal antibodies, the invention also provides various antibody fragments, such as Fab, F(ab')₂, Fv, and sFv fragments.

5 Recombinant, chimeric, and humanized antibodies are also provided.

Recombinant "humanized antibodies" which specifically bind to one of the four plexin polypeptides can be synthesized according to methods known in the art (see, *e.g.*, Green L.L. *et al.*, *Nature Genetics* 7: 13-21, 1994 for fully humanized antibodies expressed in transgenic animals; see also U.S. Patent Nos. 5,693,761; 5,777,085; and 5,585,089). Humanized antibodies are chimeras comprising mostly human IgG
10 sequences into which at least portions of the regions responsible for specific antigen-binding (*e.g.*, CDR's) have been inserted. Animals are immunized with the desired antigen, the corresponding antibodies are isolated, and the portion of the variable region sequences responsible for specific antigen binding are removed. The animal-derived
15 antigen binding regions are then cloned into the appropriate position of human antibody genes in which the antigen binding regions have been deleted. Humanized antibodies minimize the use of heterologous (*i.e.*, inter-species) sequences in human antibodies, and thus are less likely to elicit immune responses in the treated subject (see also, *e.g.*, U.S. Patent No. 5,807,715).

20 Construction of different classes of recombinant antibodies can also be accomplished by making chimeric or humanized antibodies comprising nonhuman variable domains and human constant domains (CH1, CH2, CH3) isolated from different classes of immunoglobulins. For example, antibodies with increased antigen binding site valencies can be recombinantly produced by cloning the antigen binding
25 site into vectors carrying the human chain constant regions (see, *e.g.*, Arulanandam *et al.*, *J. Exp. Med.* 177: 1439-1450, 1993).

In addition, standard recombinant DNA techniques can be used to alter the binding affinities of recombinant antibodies with their antigens by altering amino acid residues in the vicinity of the antigen binding sites. The antigen binding affinity of a
30 humanized antibody can be increased by mutagenesis based on molecular modeling (see, *e.g.*, Queen *et al.*, *Proc. Natl. Acad. Sci.* 86: 10029-10033, 1989).

Also provided in the invention are plexin polypeptide-specific single polypeptide chain antibodies (see general methods in U.S. Patent Nos. 4,946,788 and

4,704,692); single domain antibodies (Ward, E.S. et al., *Nature* **341**: 544-546, 1989); and chimeric antibodies (U.S. Patent No. 4,816,567).

Binding agents that specifically bind the plexin polypeptides of the present invention are useful, for example, in determining expression levels of the plexin polypeptides in various tissues of the body, Western blotting analysis, and immunochromatography. Particularly, binding agents that specifically bind the plexin polypeptides are useful for binding the plexin polypeptide on a cell expressing the plexin polypeptide, thereby activating the cell.

A binding agent that specifically binds one of the four plexin polypeptides, for example, is effective as an immune modulator. Additional applications include, without limitation, an injectable formulation comprising a binding agent that specifically binds one of the four plexin polypeptides that is useful to antagonize activity in a disease involving aberrant immune regulation or a disease involving aberrant cell growth.

In an eighth aspect, the invention provides an isolated antisense oligonucleotide complementary to a portion of a nucleic acid molecule encoding one of the four plexin polypeptides. In certain embodiments, hybridization of the antisense oligonucleotide to the nucleic acid molecule inhibits transcription or translation of the nucleic acid molecule.

By two nucleic acid molecules being "complementary" to one another is meant that the first nucleic acid molecule (e.g., an oligonucleotide) is able to form Watson-Crick base pair hydrogen bonds (i.e., hybridize) with the second nucleic acid molecule to form a duplex. The first nucleic acid molecule is thus a "complement" of the second nucleic acid molecule.

The antisense oligonucleotides according to the invention are complementary to a region of a nucleic acid molecule (or a region at the intron/exon boundary of DNA or RNA) that encodes one of the four plexin polypeptides. Preparation of antisense oligonucleotides is well known (see, e.g., Agrawal et al., *Trends Biotechnol.* **10**:152-158, 1992; U.S. Patent No. 5,149,798; Agrawal et al., *Proc. Natl. Acad. Sci. USA* **85**:7079-7083, 1988; Froehler, *Tetrahedron Lett.* **27**:5575-5578, 1986; and Bergot et al., *J. Chromatog.* **559**:35-42, 1992).

In a ninth aspect, the invention provides a method for identifying a nucleic acid molecule encoding one of the four plexin polypeptides, comprising contacting a pool of candidate nucleic acid molecules with a nucleic acid molecule encoding one of the four

plexin polypeptides, wherein hybridization of the nucleic acid molecule encoding one of the four plexin polypeptides under stringent conditions to a candidate nucleic acid molecule identifies the candidate nucleic acid molecule as a nucleic acid molecule that encodes one of the four plexin polypeptides. According to this aspect of the invention, 5 "hybridization" and "stringent conditions" are as defined above for the first aspect of the invention. In certain embodiments, the nucleic acid molecule encoding one of the four plexin polypeptides has a nucleic acid sequence comprising SEQ ID NOS: 1, 3, 5 or 7.

It will be understood that the isolated plexin polypeptides according to the second aspect of the invention, the plexin chimeric molecules according to the fifth aspect of the 10 invention, and binding agents that specifically bind the plexin polypeptides according to the seventh aspect of the invention, are useful as therapeutics to treat an individual suffering from, or suspected of having, a disease or disorder involving aberrant immune regulation or an individual suffering from, or suspected of having, a disease or disorder involving aberrant cell growth, particularly nerve cell growth.

15 By "disease or disorder involving aberrant immune regulation" is meant any disease or disorder in which an abnormal immune response is generated in response to either self or foreign antigens. Thus, this definition includes, without limitation, autoimmune diseases (*e.g.*, lupus, inflammatory bowel disease, or Diabetes Type 1) and immunosuppressive diseases (*e.g.*, multiple sclerosis or rheumatoid arthritis).

20 By "disease or disorder involving aberrant cell growth" is meant any disease or disorder in which an abnormal amount of cell growth is observed. "Cell growth" is defined above. Thus, diseases and disorders involving aberrant cell growth include hyperplasia, neoplasia, and cancer, as well as degenerative diseases, such as neurodegenerative diseases.

25 Preferable therapeutically useful plexin polypeptides are soluble polypeptides (*e.g.*, lacking the hydrophobic transmembrane domain of the plexin polypeptides), particularly soluble polypeptide fragments that are secreted by the cell in which the fragment was produced. In a preferred embodiment the soluble plexin polypeptides are selected from the group consisting of plexin-A-1 (Maestrini et al. 1996 *supra*), plexin-A- 30 2 (Maestrini et al. 1996 *supra*), plexin-A-3 (Maestrini et al. 1996 *supra*), plexin-A-4, plexin-B-1 (Maestrini et al. 1996, *supra*), plexin-B-2, plexin-B-3, plexin-C1 (Comeau et al. 1998 *supra*), plexin-D-1.

In a tenth aspect, the invention provides a method for diagnosing a disease involving aberrant immune regulation or a disease involving aberrant cell growth, comprising comparing the amino acid sequence of one of the four plexin polypeptides from an individual suspected of having the disease with the amino acid sequence of one of the four plexin polypeptides from an unaffected individual, wherein the presence of a difference between the two amino acid sequences identifies the individual suspected of having the disease as having the disease. "Disease or disorder involving aberrant immune regulation" and "disease or disorder involving aberrant cell growth" are as defined above.

By "difference" in the amino acid sequence of one of the four plexin polypeptides from an individual suspected of having the disease or disorder as compared with the amino acid sequence of one of the four plexin polypeptides from an unaffected individual, is meant any mutation that changes the amino acid sequence including substitution, deletion, of addition of one or more amino acid residues.

Thus, in one nonlimiting example, one of the four plexin polypeptides is extracted from cells of an individual suspected of having a disease involving aberrant immune regulation (*e.g.*, using an antibody according to the seventh aspect of the invention). The amino acid sequence of the plexin polypeptide is determined by N-terminal sequencing and compared to the amino acid sequence of one of the four plexin polypeptides from an unaffected individual (*i.e.*, a normal individual of the same species that does not have a disease involving aberrant immune regulation or a disease involving aberrant cell growth). If there is a difference in the two amino acid sequences, the individual suspected of having a disease involving aberrant immune regulation is identified as having a disease involving aberrant immune regulation, and may be treated accordingly.

In certain embodiments of the tenth aspect, the amino acid sequence of the plexin polypeptide from the unaffected individual comprises the sequence of SEQ ID NO: 2, 4, 6, 8 or 10.

The following examples are intended to further illustrate certain preferred embodiments of the invention and are not limiting in nature. Those skilled in the art will recognize, or be able to ascertain, using no more than routine experimentation, numerous equivalents to the specific substances and procedures described herein. Such

equivalents are considered to be within the scope of this invention, and are covered by the following claims.

Practice of the present invention will employ, unless indicated otherwise, conventional techniques of cell biology, cell culture, molecular biology, microbiology, recombinant DNA, protein chemistry, and immunology, which are within the skill of the art. Such techniques are described in the literature. See, for example, **Molecular Cloning: A Laboratory Manual**, 2nd edition. (Sambrook, Fritsch and Maniatis, eds.), Cold Spring Harbor Laboratory Press, 1989; **DNA Cloning**, Volumes I and II (D.N. Glover, ed), 1985; **Oligonucleotide Synthesis**, (M.J. Gait, ed.), 1984; U.S. Patent No. 4,683,195 (Mullis et al.); **Nucleic Acid Hybridization** (B.D. Hames and S.J. Higgins, eds.), 1984; **Transcription and Translation** (B.D. Hames and S.J. Higgins, eds.), 1984; **Culture of Animal Cells** (R.I. Freshney, ed). Alan R. Liss, Inc., 1987; **Immobilized Cells and Enzymes**, IRL Press, 1986; **A Practical Guide to Molecular Cloning** (B. Perbal), 1984; **Methods in Enzymology**, Volumes 154 and 155 (Wu et al., eds), Academic Press, New York; **Gene Transfer Vectors for Mammalian Cells** (J.H. Miller and M.P. Calos, eds.), 1987, Cold Spring Harbor Laboratory; **Immunochemical Methods in Cell and Molecular Biology** (Mayer and Walker, eds.), Academic Press, London, 1987; **Handbook of Experiment Immunology**, Volumes I-IV (D.M. Weir and C.C. Blackwell, eds.), 1986; **Manipulating the Mouse Embryo**, Cold Spring Harbor Laboratory Press, 1986.

The following Examples are provided to illustrate the present invention, and should not be construed as limiting thereof.

EXAMPLES

Example 1

Identification and cDNA cloning of *plexins* and sequence analysis

Since the coding sequences of human *plexin-B1(SEP)*, *plexin-A2(OCT)* and
 5 *plexin-A1(NOV)* were incomplete, we obtained the missing cDNA by RT-PCR; primers
 were designed by homology to orthologous murine sequences and corresponding ESTs.
 Updated database entries are X87904, X87831 and X87832, respectively. Partial cDNA
 of *plexin-A4* was obtained by assembling five overlapping human ESTs (HGI THC
 Report: THC203425), deriving from chromosome 7 specific cDNA pools. Another EST
 10 from chr: 7 (clone 7B19F10) encodes the cytoplasmic domain of a *plexin* and
 presumably derives from the same gene as *plexin-A4*. *Plexin-B2* cDNA was amplified
 by RT-PCR starting from the partial cDNA sequences of clones *MM1* (Shinoura, N.,
 Shamraj, O.I., Hugenholtz, H., Zhu, J.G., McBlack, P., Warnick, R., Tew, J.J., Wani,
 M.A., and Menon, A.G. (1995). Identification and partial sequence of a cDNA that is
 15 differentially expressed in human brain tumors. *Cancer Lett* 89, 215-221) and
 KIAA0315 (Genbank database); the genomic locus of *SEP-B* was identified due to its
 100% sequence identity with clone C22_311 from human chromosome 22. *Plexin-B3*
 coding sequence was identified in the genomic sequence of ALD locus on human
 chromosome Xq28, using the algorithms HEXON and GENIE. *Plexin-D1* was similarly
 20 found in the genomic sequence of chromosome 3 (pac pDJ70i11). The genomic
 sequence of *plexin-B1(SEP)*, in the region of the alternative splicing of the extracellular
 domain, was obtained using the following primers: sense
 5'GCAGCACCTGTGCACCCACAAGGC3' and antisense:
 5'TGCAGGCTGGACGGGAGGATGAGG3'. The common donor site is
 25 CCATCAG/gtgattgt (position 2028 from ATG); the alternative splice acceptor sites are:
 (i) ccccttcag/AGCCC, leading to the canonical *plexin-B1* sequence, and (ii)
 ctctctcag/GTGAT, leading to "plexin-B1 truncated" variant. All these new sequences
 were analyzed using the algorithms BLAST2, NETPHOS (phosphorylation prediction
 sites, by Nicolaj Blom), PH-PREDICT and Consensus Protein Secondary Structure
 30 prediction at IBCP. The phylogenetic tree was generated using AllAll algorithm of the
 Darwin sequence analysis system (at CBRG).

Example 2

Plexin cDNA expression constructs and protein analysis

Cell transfections were carried out by Calcium phosphate and DEAE-dextran methods, using 5-10 µg of each cDNA (1-2 µg each in case of cotransfections). For transient transfections in COS and BOSC-23 cells the cDNA was cloned in pCDNA3 or derived expression plasmids (Invitrogen). MDCK stable transfectants for *plexin-A3* were obtained using pCEP4 expression plasmid (Invitrogen); the selection was done in the presence of Hygromycin-B (100-200 µg/ml). *Plexin-A3* positive clones of MDCK cells were isolated from two independent transfections, and showed identical biological properties. *Plexin* and neuropilin expression constructs included a VSV- and myc-tag at the N'- and C' protein-termini, respectively, detected by monoclonal antibodies anti-VSV-G (cat. V-5507, Sigma) and anti-cMyc-tag (cat. OP10-100UG, Calbiochem). "Plexin-B1 truncated" splice variant was expressed from a cDNA fragment isolated by RT-PCR and VSV-tagged at the N' terminus: the encoded amino acid sequence spans up to aa 676 (including the *sema* domain and two MRS motifs). "Plexin-B1-sema" derives from a further deletion of the *plexin-B1* extracellular domain, and exclusively includes the *sema* domain. "Plexin-B1-Δsema" protein mutant includes only the C' terminal half of *plexin-B1* extracellular domain, starting from amino acid 606, i.e. excluding *sema* domain and first MRS but including second and third MRS, transmembrane and intracellular domains.

For immunoprecipitations, cells were lysed with EB buffer (20 mM Tris-HCl pH 7.4, 5 mM EDTA, 150 mM NaCl, 10% glycerol, 1% Triton X-100), in the presence of a cocktail of protease inhibitors and 1mM Na-ortovanadate. Immunoprecipitations were performed at 4°C for 4h with the appropriate antibodies; high stringency washes were performed, in the presence of 1 M LiCl.

For *in vitro* kinase assays, immunopurified proteins were incubated with kinase buffer (50 mM Hepes, 100 µM DTT, 5 mM MnCl₂, 5 mM MgCl₂) in the presence of redivue 5 µCi [γ -³²P] ATP (Amersham) for 10 minutes at 4°C in agitation. Samples were then submitted to SDS-PAGE and autoradiography, or analysed using a Phosphor-Imager system (Molecular Dynamics). Alkali treatment of the polyacrilamide gels was performed with 1M KOH for two hours at 55°C.

Western blots were performed according to standard methods. Specific detection of phospho-tyrosines was done with PY20 MoAb (Trasduction laboratories). Final detection was done with ECL system (Amersham).

Example 3

Semaphorin-SEAP binding assays

Soluble forms of Semaphorin extracellular domains were expressed as chimeric molecules with placental Secreted Alkaline Phosphatase (SEAP) and harvested from the conditioned media of transiently transfected COS or BOSC-23 cells. Serum-free media were concentrated over 100 times using Centricon Plus-20 filters (Millipore) with a molecular weight cutoff of 100 kDa. The AP activity of these media was assessed as described (Flanagan, J.G. and Leder, P. (1990). The kit ligand: a cell surface molecule altered in steel mutant fibroblasts. *Cell* 63, 185-194); the specific activity of chimeric molecules was approx. 1000 U/mg. Concentrated Semaphorin-SEAP were diluted as appropriate in a HEPES buffered saline, additioned with 0.2% BSA, 0.1% NaN₃, 5 mM CaCl₂ and 1 mM MgCl₂ (HBSBA). For binding assays, COS cells transiently transfected with plexins were seeded on 48 well plates to reach confluence, and then incubated with Semaphorin-SEAP preparations (approx 1-5 nM) for 90 minutes at room temperature. The binding was detected as described (Flanagan and Leder, 1990). Binding experiments with plexin-C1/VESPR were as described (Comeau, M.R., Johnson, R., DuBose, R.F., Petersen, M., Gearing, P., VandenBos, T., Park, L., Farrah, T., Buller, R.M., Cohen, J.I., Strockbine, L.D., Rauch, C., and Spriggs, M.K. (1998). A poxvirus-encoded semaphorin induces cytokine production from monocytes and binds to a novel cellular semaphorin receptor, VESPR. *Immunity*. 8, 473-482; He, Z. and Tessier-Lavigne, M. (1997). Neuropilin is a receptor for the axonal chemorepellent Semaphorin III. *Cell* 90, 739-751).

For *in vitro* binding assays, plexin-B1 was purified from cell extracts by immunoprecipitation with anti-VSV antibody. Extracts of mock-transfected cells were used as control samples. After washing, the immunocomplexes were incubated with serial dilutions of CD100-SEAP (prepared as above) for 2 hours at 4°C, in continuous agitation. Samples were then washed 3 times with HBSBA and the bound alkaline phosphatase activity was measured by colorimetric assay using p-nitro-phenyl-phosphate, as described (Flanagan and Leder, 1990). Scatchard analysis was done using Equilibrate (by GertJan C. Veenstra).

Example 4

In situ hybridization analysis

RNA *in situ* hybridization was performed essentially as described (He, Z. and Tessier-Lavigne, M. (1997). Neuropilin is a receptor for the axonal chemorepellent Semaphorin III. *Cell* 90, 739-751). Briefly cDNA fragments of plexin-A1, -A2, and -A3 were used to generate ³⁵S-labeled antisense and sense RNA probes, which were used for *in situ* hybridization histochemistry of cryostat sections of rat embryos.

Example 5

Xenopus turning assay

The methods for injecting mRNA encoding various constructs, and for studying the turning responses of the neurons, are exactly as described previously (Hong, K., Hinck, L., Nishiyama, M., Poo, M.M., Tessier-Lavigne, M., and Stein, E. (1999). A ligand-gated association between cytoplasmic domains of UNC5 and DCC family receptors converts netrin-induced growth cone attraction to repulsion. *Cell* 97, 927-941; Song, H., Ming, G., He, Z., Lehmann, M., Tessier-Lavigne, M., and Poo, M. (1998). Conversion of neuronal growth cone responses from repulsion to attraction by cyclic nucleotides [see comments]. *Science* 281, 1515-1518).

Example 6

Mixed-culture assays and time-lapse videomicroscopy

Mock-transfected and plexin-A3 overexpressing MDCK cells were seeded with mesenchymal cells (NIH 3T3, KJ29, D17, among others), in multiwell culture plates by 1:4 or 1:1 ratio. NIH and KJ-29 cells were sometimes labeled by addition of DiI (1,1'-diiododecyl-3,3,3',3'-tetramethylindocarbocyanine perchlorate, Fluka) in the culture medium, 4 hours before harvesting for the assay; clusters of cells marked with this dye are marked in blue (in light microscopy) and emit red epifluorescence (TRITC filter). The repelling effect was observed 16-30 hours after confluency, by contrast phase microscopy using Leica DM IL. The progress of the assays was also monitored by time-lapse video-microscopy (320 minutes recording were converted into 1 minute play). To determine the time-length of cell contacts, for each assay, randomly chosen fibroblasts were followed during several hours and the duration of each contact between their lamellipodia and MDCK cells was measured. The doubling time of cells and their viability during the assay could also be analyzed, and no differences were observed in presence of control or plexin-A3 expressing cells. Substrate adhesion of plexin-A3 overexpressing MDCKs was analyzed by counting attached cells after 30 minutes from

seeding on micro-wells coated with fibronectin, collagen or polylysine, in the absence of calf serum: no differences versus control cells were observed.

Example 7

Apoptosis detection

5 TUNEL reaction (Boehringer detection kit) was performed on mixed cultures of MDCK and NIH3T3 cells, 24 hours after seeding in a 24-well culture plate. The labeling was converted into a colorimetric signal for analysis by light microscopy using the TUNEL-AP detection kit (Boehringer). As a positive control for the induction of apoptosis, the same cells were treated with UV-C ($50 \mu\text{J}/\text{cm}^2$) or $1 \mu\text{M}$ staurosporin.

10 Although the foregoing invention has been described in some detail by way of illustration and example for purposes of clarity of understanding, it will be apparent to those skilled in the art that certain changes and modifications will be practiced. Therefore, the description and examples should not be construed as limiting the scope of the invention, which is delineated by the appended claims.

15 Example 8

Plexins are specific receptors for cell surface semaphorins in vertebrates

Plexin-C1 (VESPR) has been shown to bind the soluble viral semaphorins Sema VA and VB (Comeau et al., 1998 *supra*), and we recently found that *Drosophila* Plexin A (D Plex A) interacts with transmembrane Sema 1a (Winberg et al., 1998 *supra*). We
20 therefore examined in vertebrates whether the extracellular domain of several different cellular semaphorins -fused to alkaline phosphatase- could bind members of the human plexin-A, -B and -C subfamilies. Multiple secreted semaphorins of class 3 (Sema3A, Sema 3C or Sema3F; see below) did not interact with plexins-A1, -A2, -A3, -B1, B2, or -C1 (data not shown). In contrast, plexin-C1(Vespr) specifically bound Sema7A(Sema-
25 K1) (Fig. 2a), a GPI-membrane linked semaphorin (class 7). This result is not entirely unexpected, since Sema7A may represent the cellular counterpart of viral semaphorin SemaVB, previously shown to interact with this plexin (Comeau et al., 1998 *supra*). More interestingly, the class 4 transmembrane semaphorin Sema4D (CD100) did
30 interact strongly and specifically with plexin-B1 (Fig. 2a). Thus the prototypes of two distinct plexin families are the receptors for members of two distinct semaphorin sub-classes. We also found that Sema7A and Sema4D do not bind to neuropilin-1 or -2 alone, nor did co-transfection of either neuropilin with plexin-B1 significantly modify

its binding efficiency (not shown). Neuropilins thus seem so far to function as receptors only for vertebrate semaphorins of class 3.

The affinity constant of Sema4D for plexin-B1 was estimated by Scatchard plot to be in the subnanomolar range ($K_D = 0.9$ nM, Fig. 2b; the estimated K_D of Sema7A for plexin-C1 is 2.1 nM, not shown). These values are consistent with those observed for semaphorins-neuropilins, and fly semaphorin1-Plexin A interactions (He and Tessier-Lavigne, 1997 *supra*; Winberg et al., 1998).

We used two deletion constructs of plexin-B1 to explore the semaphorin binding sites of plexins. Neither the N-terminal half of plexin-B1 extracellular domain ("plexin-B1-truncated", see previous paragraph), nor its C-terminal half ("plexin-B1- Δ sema", see Experimental Procedures) was sufficient alone to bind CD100 (see Fig. 2a), suggesting that the binding of Sema4D depends on multiple structural determinants of the extracellular domain of plexin-B1.

Example 9

Plexins associate with class 3 Semaphorin receptors, Neuropilins

As outlined above, secreted semaphorins of subclass 3 are known to bind neuropilins (He and Tessier-Lavigne, 1997 *supra*; Kolodkin et al., 1997 *supra*; Chen, H., Chedotal, A., He, Z., Goodman, C.S., and Tessier-Lavigne, M. (1997) "Neuropilin-2, a novel member of the neuropilin family, is a high affinity receptor for the semaphorins Sema E and Sema IV but not Sema III." *Neuron* 19, 547-559). However, the short cytoplasmic tail of neuropilins seems to be dispensable for their biological activity (Nakamura, et al (1998) *supra*), indicating the requirement of an associated co-receptor for signal transduction. Interestingly, in *Drosophila* (where neuropilins have not been identified to date) Plexin A is sufficient to mediate the biological response to semaphorin-1 in axon guidance (Winberg et al., 1998 *supra*).

In an initial set of experiments, we could not observe binding of the class 3 semaphorins Sema3A(Sema III), Sema3C(Sema E) or Sema3F(Sema IV) to plexins-A1, A2, A3, B1, B2 or C1 (not shown). To test whether plexins might be coreceptors with neuropilins for class 3 semaphorins, we set up co-precipitation experiments in COS cells to test whether neuropilins may interact with plexins. Three tested plexins (plexin-A1, -A3 and -B1) associated both with neuropilin-2 (Np2, shown in Fig. 3) and neuropilin-1 (not shown). The binding was specific, inasmuch as neither neuropilin nor any plexins coimmunoprecipitated with the netrin receptor DCC, under conditions

where DCC coimmunoprecipitated with the other netrin receptor UNC5H2 (Fig. 3 and data not shown). We observed finally that the plexin-neuropilin association is mediated by the *sema domain* of plexins, as demonstrated using either the "plexin-B1 truncated" splice variant (Figure 3) or an even shorter form of the extracellular domain ("plexin-B1-sema", see Experimental procedures, not shown).

To further support the idea of a plexin-neuropilin multimeric receptor complex for semaphorins, we show here that plexin-A3 (e.g.) is expressed in a large number of neuronal classes, including sensory, sympathetic, motor, and olfactory bulb neurons (Figure 4 and data not shown), which are known to respond to class 3 semaphorins, and which express either neuropilin-1 or neuropilin-2 or both (Chen et al., 1997 *supra*; Feiner, L., Koppel, A.M., Kobayashi, H., and Raper, J.A. (1997). Secreted chick semaphorins bind recombinant neuropilin with similar affinities but bind different subsets of neurons in situ. *Neuron* 19, 539-545; He and Tessier-Lavigne, 1997 *supra*; Kolodkin et al., 1997 *supra*). Thus, plexin-A3 is a candidate for a physiological coreceptor involved in mediating class 3 semaphorin effects on these axons. Other plexins may also have a role as neuropilin coreceptors in specific cell populations, such as plexin-A2, which is expressed in a subset of sensory neurons and in dorsal horn cells, and plexin-A1, which is expressed at low levels and broadly in the spinal cord (Figure 4).

To directly test the possible involvement of plexins in class 3 semaphorin signal transduction, we studied the repulsive responses of *Xenopus* spinal neurons to Sema3A, which is mediated by a receptor mechanism involving neuropilin-1 (Song et al., 1998 *supra*). We asked whether these responses could be altered by expression of a presumed dominant-negative plexin-A1 construct lacking the cytoplasmic domain of the protein. Transmembrane proteins can be reliably expressed in these neurons by injecting the encoding mRNA at the developmental two cell stage, allowing the embryos to grow to tadpole stage, and then removing the spinal cord and culturing the neurons (Hong et al., 1999 *supra*). We therefore injected the mRNA encoding the truncated plexin-A1 construct, together with mRNA encoding GFP (as a reporter) and then studied the responses of spinal neurons expressing GFP that were derived from these embryos. Whereas control spinal neurons are repelled by Sema3A (Figure 5A, B and Song et al. 1998 *supra*), neurons from embryos injected with mRNA for truncated plexin-A1 did not respond with either repulsion or attraction to Sema3A (Figure 5C,

D). This blocking effect appeared to be specific, since expression of a different heterologous receptor, UNC5H2, did not impair repulsion by Sema3A (Hong et al., 1997 *supra*), and since expression of the truncated plexin construct did not block attractive responses to netrin-1 (Figure 5E, F). Figure 5G, H quantifies these effects.

- 5 As can be seen, the effect of Sema3A is completely abolished by the truncated plexin; although there is a slight apparent decrease in the attractive effect of netrin-1 the effect is not statistically significant.

Although we have used a truncated plexin-A1 construct, this construct may be expected to interfere with the function of various plexins, since all the plexins tested
10 (A1, A3 and B1) associated with neuropilin-1. These results support a role for one or more plexins in mediating the repulsive Sema3A signal in the *Xenopus* spinal neurons.

Example 10

Plexins signal via a novel type of tyrosine phosphorylated cytoplasmic domain

The sequences of plexin cytoplasmic domains are highly conserved among
15 plexins but do not match any known sequences. We found that the plexin-A3 and plexin-B1 proteins are phosphorylated on tyrosine residues when overexpressed in human kidney cells (BOSC-23), as demonstrated using anti-phosphotyrosine antibodies (Fig. 6a). Furthermore, after immunoprecipitation and in vitro kinase assays, plexin-A3 and plexin-B1 became phosphorylated (Fig. 6b). Resistance to an alkali treatment (see
20 Experimental procedures) confirmed the specific phosphorylation of tyrosine residues.

The cytoplasmic domains of several receptors, including Met proteins, become tyrosine phosphorylated owing to an intrinsic kinase activity (Ullrich, A. and Schlessinger, J. (1990) "Signal transduction by receptors with tyrosine kinase activity." *Cell* 61, 203-212). Since the cytoplasmic domain of plexins is not similar to any bona
25 fide or atypical tyrosine kinase, this suggests that a distinct tyrosine kinase co-immunoprecipitates in association with plexins, and is responsible for their tyrosine phosphorylation. Although some additional phosphorylated proteins can be found specifically with plexin-A3 and -B1, we have not as yet identified this associated kinase. A number of endogenously expressed tyrosine kinases, namely Met, Ron, Abl
30 and Src, were not found associated with plexin-A3 by immunoprecipitation and Western blotting (not shown). Since tyrosine phosphorylated residues often function as docking sites for intracellular signal transducers (Cantley, L.C., Auger, K.R., Carpenter, C., Duckworth, B., Graziani, A., Kapeller, R., and Soltoff, S. (1991) "Oncogenes and

signal transduction." Cell 64, 281-302), the fact that the cytoplasmic domains of plexins are tyrosine phosphorylated further suggests that they are part of signaling complexes.

Example 11

5 Plexin-A3 expressing cells induce repulsion of co-cultured cells

Stable transfectants expressing recombinant human plexin-A3 were successfully obtained in four different cell lines: IMR32 and AF8 (human neuroblasts), and BOSC-23 and MDCK (human and canine kidney cells, respectively). We observed modest phenotypic changes in the transfected cells, which generally become flatter and larger in
10 size. The growth rate of plexin-A3 overexpressing cells was comparable to parental lines and we did not observe differences in the ability to adhere on different substrates (data not shown).

In keeping with previous report on the related Plexin of *Xenopus laevis* (Ohta, K., Mizutani, A., Kawakami, A., Murakami, Y., Kasuya, Y., Takagi, S., Tanaka, H.,
15 and Fujisawa, H. (1995). "Plexin: a novel neuronal cell surface molecule that mediates cell adhesion via a homophilic binding mechanism in the presence of calcium ions." Neuron 14, 1189-1199), we observed a modest increase in calcium-dependent homotypic cell aggregation of plexin-A3 transfectants (not shown). Surprisingly, we found that epithelial MDCK cells overexpressing plexin-A3 mediate strong repelling
20 cues for adjacent cells. This was observed by co-culturing mock-transfected and plexin-A3 overexpressing MDCK cells together with several non-epithelial cell lines (such as NIH3T3, KJ29, and D17; Fig. 7A). Mock MDCKs grew alongside mesenchymal cells until confluency, when both cell types stopped proliferating. In contrast, when plexin-A3-overexpressing epithelial cells were grown in the same conditions, the adjacent
25 mesenchymal cells withdrew from them, and ultimately detached from the plate.

To analyze the dynamics of this repulsion process, we monitored for 36 hours, by time-lapse video-microscopy, mixed cultures of transfected MDCK cells and fibroblasts, in a number of independent experiments. At low cell density, fibroblasts showed intrinsic motility, exploring the surface of the plate with long lamellipodia and
30 filopodia, and thus coming in contacts with a high number of stationary MDCK islets. The time-length of the contacts between fibroblasts and control MDCK cells varied from 30 minutes to several hours, lasting mostly over 100 minutes. However, when fibroblasts were cultured with MDCK cells overexpressing plexin-A3, transient

contacts were observed, often lasting less than 30 minutes (see Fig. 7C). At higher cell density, fibroblasts stopped and clustered alongside the islands of control MDCKs, whereas they kept moving in a hectic fashion between the islands of plexin-A3 transfected cells (data not shown).

5 This cell-repelling effect is not due to the release of soluble factors, since exchanging conditioned media between mixed cultures was without effect (not shown). Moreover, the two different cell populations grew normally until they came into contact, indicating that the repelling effect requires cell-cell interaction. To rule out the possibility that plexin-A3 expressing cells generate an apoptotic signal for fibroblasts,
10 we monitored cell viability and apoptosis by TUNEL staining. As shown in Figure 7B, the clusters of repelled fibroblasts did not include apoptotic cells; furthermore, the detaching cells still excluded Trypan blue stain and were able to spread again on a new culture plate (not shown).

 Taken together, these results demonstrate that in our experimental system, plexin-
15 A3 mediates cell repelling cues, presumably by interacting with surface bound ligands on opposing cells. We could not identify –so far– the specific ligand for plexin-A3, however we propose that this may be a transmembrane semaphorin. It should be noted that the intracellular domains of transmembrane semaphorins, such as Sema4D, also include tyrosine residues, which may themselves become phosphorylated and associate
20 with cytoplasmic signal transducer molecules, a property shown for ligands of the ephrin family (Holland, S.J., Gale, N.W., Mbamalu, G., Yancopoulos, G.D., Henkemeyer, M., and Pawson, T. (1996) "Bidirectional signalling through the EPH-family receptor Nuk and its transmembrane ligands." *Nature* 383, 722-725).

What is claimed is:

- 5 1. Isolated nucleic acid having at least 80% nucleic acid sequence identity to a nucleotide sequence that encodes an amino acid sequence selected from the group consisting of the amino acid sequence shown in (SEQ ID NO: 2 (plexin B-2)), (SEQ ID NO: 4 (plexin B-3)), (SEQ ID NO: 6 (plexin D-1)) and (SEQ ID NO: 8 (plexin A-4))
- 10 2. Isolated nucleic acid having at least 80% nucleic acid sequence identity to a nucleotide sequence selected from the group consisting of the nucleotide sequence shown (SEQ ID NO: 1 (plexin B-2)), (SEQ ID NO: 3 (plexin B-3)), (SEQ ID NO: 5 (plexin D-1)) and (SEQ ID NO: 7 (plexin A-4)).
3. A vector comprising the nucleic acid of any one of claims 1 or 2.
- 15 4. An isolated polypeptide having at least 80% amino acid sequence identity to an amino acid sequence selected from the group consisting of the amino acid sequence shown in (SEQ ID NO: 2 (plexin B-2)), (SEQ ID NO: 4 (plexin B-3)), (SEQ ID NO: 6 (plexin D-1)) and (SEQ ID NO: 8 (plexin A-4)).
5. An isolated polypeptide having at least 80% amino acid sequence identity to:
 - 20 (a) the polypeptide shown in (SEQ ID NO: 2 (plexin B-2)), (SEQ ID NO: 4 (plexin B-3)), (SEQ ID NO: 6 (plexin D-1)) and (SEQ ID NO: 8 (plexin A-4)), lacking its associated signal peptide;
 - (b) an extracellular domain of the polypeptide shown in (SEQ ID NO: 2 (plexin B-2)), (SEQ ID NO: 4 (plexin B-3)), (SEQ ID NO: 6 (plexin D-1)) and (SEQ ID NO: 8 (plexin A-4)), with its associated signal peptide; or
 - 25 (c) an extracellular domain of the polypeptide shown in (SEQ ID NO: 2 (plexin B-2)), (SEQ ID NO: 4 (plexin B-3)), (SEQ ID NO: 6 (plexin D-1)) and (SEQ ID NO: 8 (plexin A-4)), lacking its associated signal peptide.

6. A chimeric molecule comprising a polypeptide according to claim 4 or 5 fused to a heterologous amino acid sequence.
7. The chimeric molecule of claim 6, wherein the heterologous amino acid sequence is a Fc region of an immunoglobulin.
- 5 8. An antibody that specifically binds to a polypeptide according to claim 4 or 5.
9. The antibody according to claim 8, wherein the antibody is a monoclonal, a humanized antibody or a single-chain antibody.
- 10 10. A method of suppressing or altering aberrant cell growth involving a signaling pathway between a plexin and a neuropilin in a mammal comprising the step of administering an effective amount of an agent to said mammal capable of interfering with the association between the plexin and neuropilin
- 15 11. A method of treating, suppressing or altering a disorder involving aberrant immune regulation involving a signaling pathway between a plexin and a neuropilin in a mammal comprising the step of administering an effective amount of an agent to said mammal capable of interfering with the association between the plexin and neuropilin.
- 20 12. The method according to claim 10 or 11 wherein said agent is a chimeric molecule according to claim 6 or 7.
13. The method according to claim 10 or 11, wherein said agent is an antibody according to claim 8 or 9.
- 25 14. A method of diagnosing or screening for tumors in a subject characterized by the expression profiles of the polypeptides according to claim 4 or 5 wherein the expression profile of the polypeptides is different in a non-tumor sample as compared to the expression profile of the polypeptides in a tumor sample.

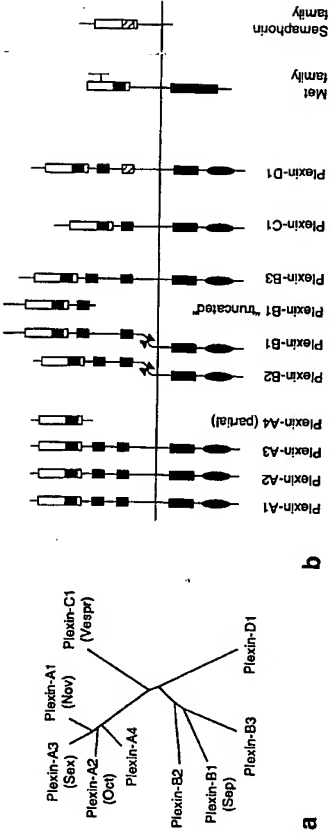
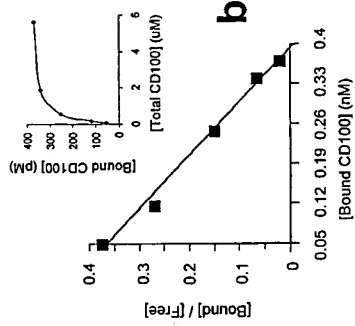
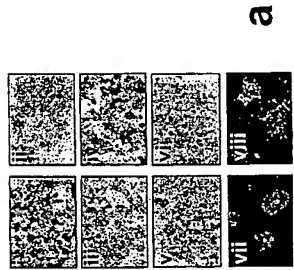
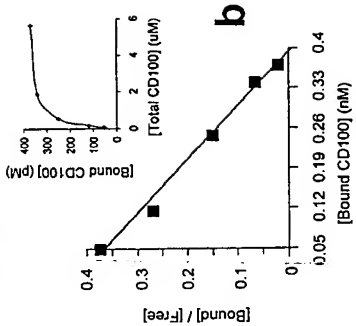


Figure 1 (Tamagnone et al.)



F16.2

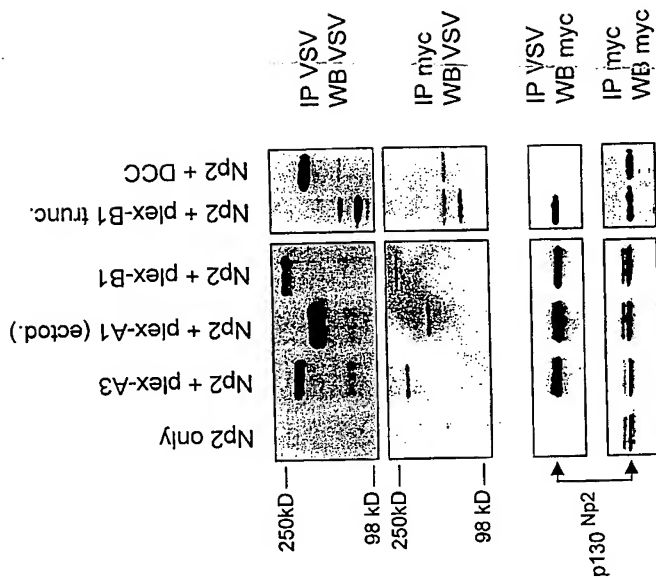


FIG. 3

4/7

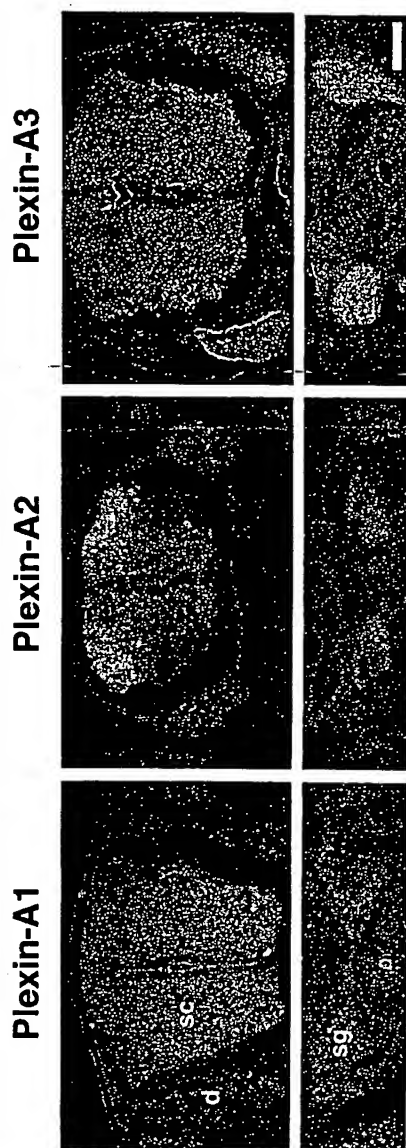


FIG. 4

5/7

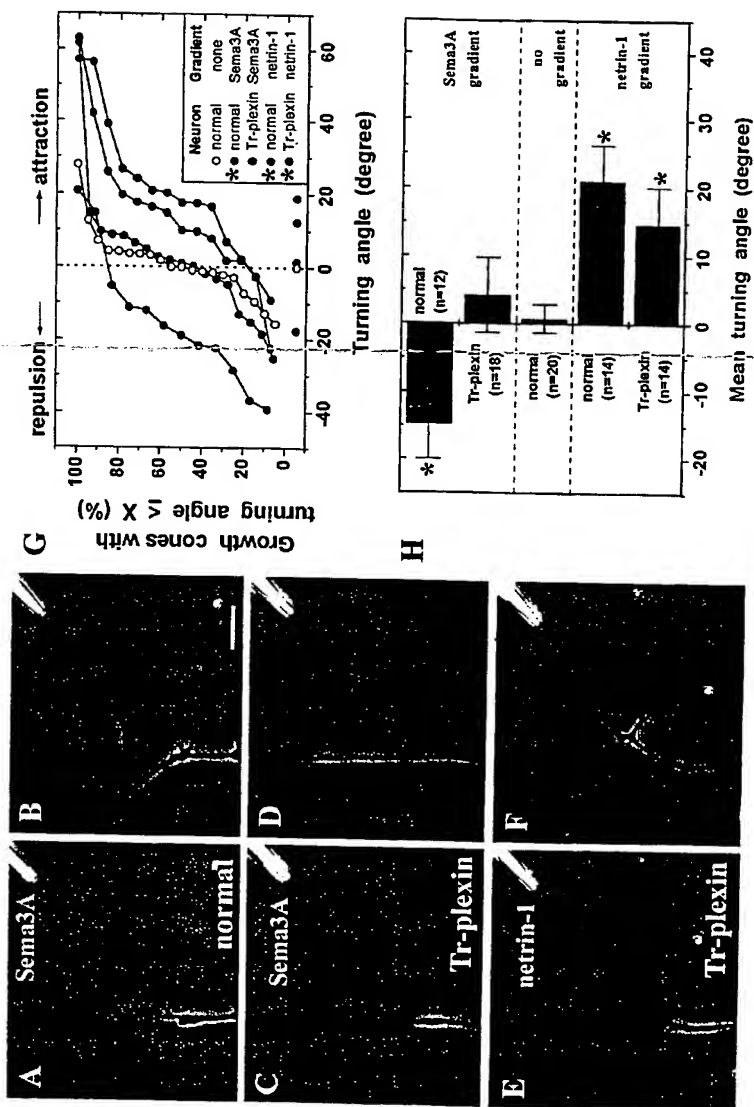
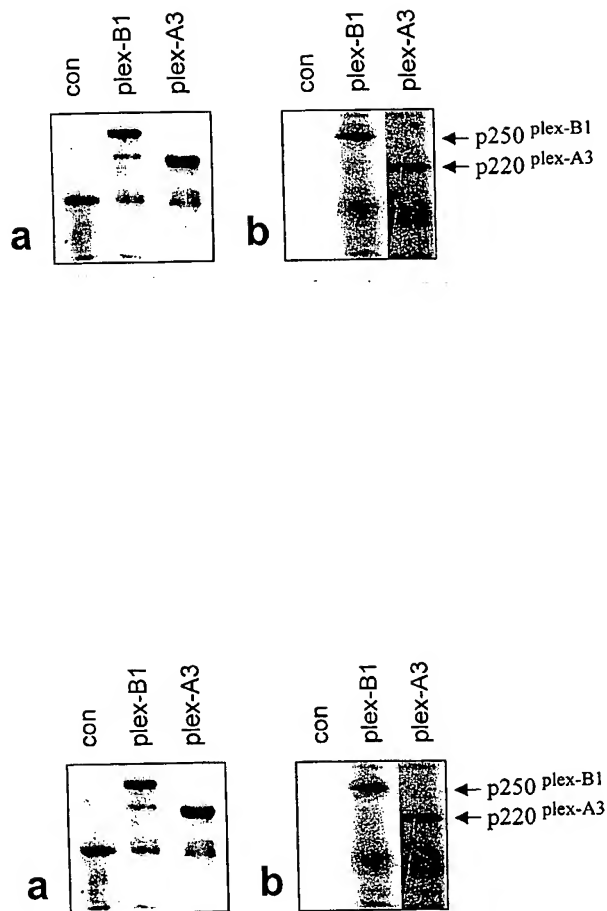
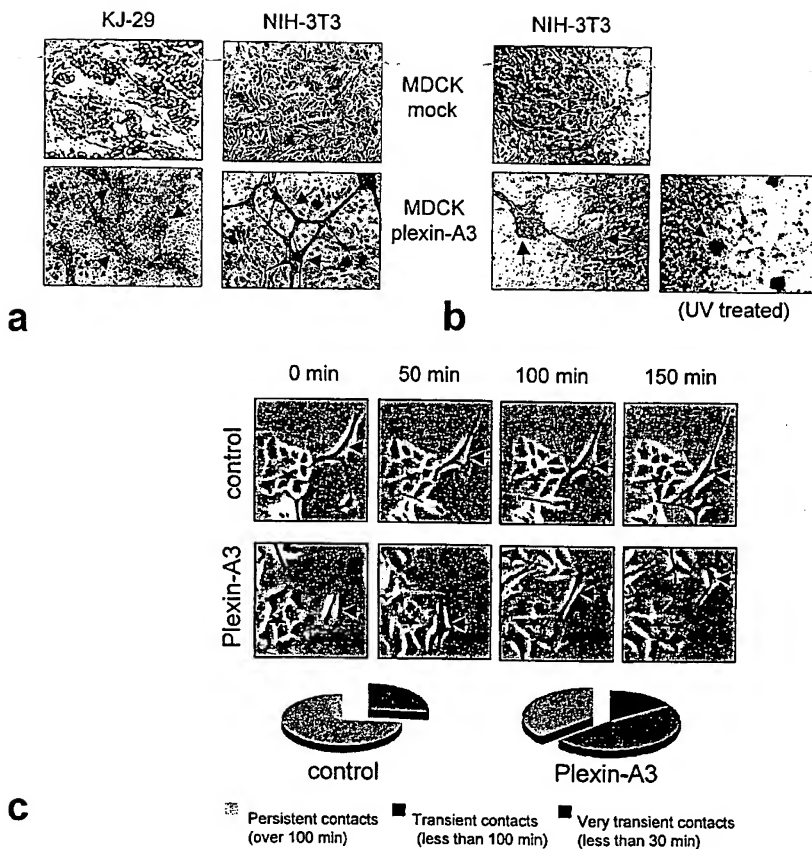


FIG. 5



7/7
FIG. 7



SEQUENCE LISTING

<110> University of Torino

<120> Novel Plexins and Uses Thereof

<130> A077PCT

<140> Not assigned yet

<141> 2000-08-25

<150> 60/150576

<151> 1999-08-25

<160> 12

<170> FastSEQ for Windows Version 4.0

<210> 1

<211> -6252

<212> DNA

<213> HOMO SAPIEN

<400> 1

gcgggggggca	atggcactgc	agctctgggc	cctgaccctg	ctgggcctgc	tggggcgagg	60
tgccagccctg	aggccccgca	agctggactt	cttccgcagc	gagaaagagc	tgaaccacct	120
ggctgtggat	gaggcctcag	gcgtgggtga	cctgggggcg	gtgaatgcc	tctaccagct	180
ggatgcgaag	ctgcagctgg	agcagcaggt	ggccacgggc	ccggccctgg	acaacaagaa	240
gtgcacgccg	ccatcgagg	ccagccagtg	ccatgaggct	gagatgactg	acaatgtcaa	300
ccagctgctg	ctgctcgacc	ctcccaggaa	gcgcctgggt	gagtgcgga	gcctcttcaa	360
gggcatctgc	gctctgcgcg	ccctgagcaa	catctccctc	cgctgttct	acgaggacgg	420
cagcggggag	aagtcttttcg	tggccagcaa	tgatgagggc	gtggccacag	tggggctggt	480
gagctccacg	ggctctgggt	gtgaccgcgt	gctgtttgtg	ggcaaaaggca	atgggccaca	540
cgacaacggc	atcatcgtga	gcactcggct	gttggaccgg	actgacagca	gggaggcctt	600
tgaagcctac	acggaccacg	ccactacaa	ggccggctac	ctgtccacca	acacacagca	660
gttcgtggcg	gccttcgagg	acggccccta	cgtcttcttt	gtcttcaacc	agcaggacaa	720
gcacccggcc	cggaaaccga	cgtctgtggc	acgcatgtgc	agagaagacc	ccaactacta	780
ctcctacctg	gagatggacc	tgcagtgcgc	ggaccccgac	atccacggcg	ctgcctttgg	840
cacctgcctg	gccgcctccg	tggctgcgcc	tggctctggc	agggtgctat	atgctgtctt	900
cagcagagac	agccggagca	gtggggggcc	cggtgccggc	ctctgcctgt	tcccgctgga	960
caaggtgcac	gccaaagatg	aggccaaccg	caacgcctgt	tacacaggca	cccgggaggg	1020
ccgtgacatc	tctacaaagc	ccttccacgg	cgatatccag	tgcggcgggc	acgcgcgggg	1080
ctccagcaag	agcttcccat	gtggctcgga	gcacctgcc	taccgcctgg	gcagccgcga	1140
cgggctcaga	ggcacagccg	tgctgcagcg	tggaggcctg	aacctcacgg	ccgtgacggg	1200
cgccgcggag	aacaaccaca	ctgttgcctt	tctgggcacc	tctgatggcc	ggatcctcaa	1260
ggtgtacctc	accccagatg	gcacctcctc	agagtacgac	tctatccttg	tggagataaa	1320
caagagagtc	aagcgcgacc	tggactgtgc	tggagacctg	ggcagcctgt	acgccatgac	1380
ccaggacaag	gtgttccggc	tgcgggtgca	ggagtgcctg	agctaccgca	cctgcacca	1440
gtgcccgac	tcccaggacc	cctactgcgg	ctggtgcgtc	gtcgagggac	gatgcaccgg	1500
gaaggccgag	tgtccggggg	ccgaggaggc	cagccactgg	ctgtggagcc	gaagcaagtc	1560
ctgcgtggcc	ctcaccagcg	cccagccaca	gaacatgagc	cggcgggccc	agggggagggt	1620
gcagctgacc	gtcagccccc	tccctgcctt	gagcgaggag	gacgagttgc	tgtgcctttt	1680
tgggggagtcg	ccgccacacc	cgcgcccgct	ggagggcgag	gccgtcatct	gcaactcccc	1740

aagcagcatc	cccgtcacac	cgccaggcca	ggaccacgtg	gccgtgacca	tccagctcct	1800
ccttagacga	ggcaacatct	tcctcacgtc	ctaccagtag	cccttctacg	actgcccgca	1860
ggccatgagc	ctggaggaga	acctgcccgtg	catctcctgc	gtgagcaacc	gctggacctg	1920
ccagtgggac	ctgcgttacc	cagagtgcgc	ggaggcttcg	cccaaccttg	aggacggcat	1980
gtcccggtcc	cacatggagg	acagctgtcc	ccagttctcg	ggaccagacc	ccctgggtgat	2040
ccccaagaa	cacgagacag	atgtgaactt	ccaggggcaag	aacctgggaa	ccgtgaagg	2100
ttcctccctg	cacgtgggca	gtgacttgct	caagttcatg	gagccggtga	ccatgcagga	2160
atctgggacc	ttcgcttttc	ggaccccaaa	gctgtcccac	gatgccaaag	agacgctgcc	2220
cccgccacct	tacgtcaagt	cttacggcaa	gaatatcgac	agcaagctcc	atgtgacctc	2280
ctacaactgc	tcttttggcg	gcagcgactg	cagcctgtgc	cgggccccta	accccagcta	2340
caggtgtcgc	tgtgtcgggg	gccagagcag	gtgcgtgtaa	gaggccctgt	gcaacaccac	2400
ctccgagtag	ccgcccgcgc	tcatcaccag	gatccagcct	gagacggggc	ccctgggtgg	2460
gggcaaccgc	atcaccatcc	tgggggtcaa	tttgggcgtc	caagcagggg	acatccagag	2520
gatctctgtg	gccggccgga	actgtcctct	tcagccggaa	cgttactccg	tgtccaccgc	2580
gacgcgtgtg	gtgatcgagg	ctgcggagac	gcctttcacg	gggggtgtcg	aggtggacgt	2640
ctccggaaaa	ctgggcccgt	cgccctccaa	tgtccagttc	accttccaa	agcccaagcc	2700
tctcagctgtg	gagcccgagc	agggaccgca	ggcgggcccgc	accacactga	ccatccacgg	2760
caccacactg	gacacgggct	ccacaggagg	cgtaggggtg	accttcaacg	gcgtcccctg	2820
taagtgacg	aagtttgggg	cgacgctcca	gtgtgtcact	ggcccccgag	cgacacgggg	2880
ccagatgctt	ctggaggtct	cctacggggg	gtcccccgctg	cccaaccccc	gcatcttctt	2940
cacctaccgc	gaaaaccccc	tactgcgagc	cttcgagccg	ctacgaagct	ttgccagttg	3000
tggccgcgct	atcaaggtca	cgggtcaggg	cttcagcctg	atccagaggt	ttgccatggt	3060
ggtctatcgc	gagccccctg	agtcttgcca	gccgccgctg	gaggctgaat	ccctgcagcc	3120
catgacggtg	gtgggtagtc	actacgtgtt	ccacaatgac	accaaggtcg	tcttctgttc	3180
cccggctgtg	cctgaggagc	cagagggcta	caacctcacg	gtgctgacgc	agatggacgg	3240
gcaccgtgcc	ctgctcagaa	cagagggccg	ggccttcgag	tacgtgcctg	acccccacct	3300
tgagaacttc	acaggttgccg	tcaagaagca	ggtcaacaag	ctcatccagc	ccccggggac	3360
caatctgaac	aaggcgatga	cgctgcagga	ggccgagggc	tctgtgggtg	ccgagcgctg	3420
caccatgaag	acgctgaacg	agaccgacct	gtactgtgag	ccccgggag	tgcagcccc	3480
gcccaagcgg	cggcagaaaa	gagacaccac	acacaacctg	cccaggttca	ttgtgaagtt	3540
cggctctcgc	gagtgggtgc	tgggcgcgct	ggagtacgac	acacgggtga	gcgacgtgcc	3600
gcctcagcct	atcttgccgc	tgggtcatcg	ggccatggtg	gtcgtcatcg	cggtgtctgt	3660
ctactgctac	tggaggaaga	gccagcaggg	cgaacgagag	tatgagaaga	tcaagtccca	3720
gctggagggc	ctggaggaga	gcgtgcggga	ccgctgcaag	aaggaattca	cagacctgat	3780
gatcgagatg	gaggaccaga	ccaacgacgt	gcacgagggc	ggcatccccg	tgcctggacta	3840
caagacctac	accgaccggc	tcttcttctt	gccctccaag	gacggcgaca	aggacgtgat	3900
gatcaccggc	aagctggaca	tcctctgagc	gcggcgcccg	gtgtggagag	agggcctcta	3960
ccagttctcc	aacctgctga	acagcaagtc	ttctctcttc	aatttcatcc	acacctggga	4020
gaaccagcgg	gagttctcgg	cccgcgcgca	ggcttacttc	gcgtcccctg	tgacggtggc	4080
gctgcacggg	aaactggagt	actacacgga	catcatgcac	acgctcttcc	tggagctcct	4140
ggagcagtag	gtgggtggcca	agaaccccaa	gctgatgctg	cgcaggtctg	agactgtggt	4200
ggagagagatg	ctgtccaact	ggatgtccat	ctgcctgtac	cagtacctca	aggacagtgc	4260
cggggagccc	ctgtacaagc	tcttcaaggc	catcaaacat	caggtggaaa	agggcccggt	4320
ggatgcggta	cagaagaagg	ccaagtacac	tctcaacgac	acggggctgc	tgggggatga	4380
tgtggagtag	gcacccctga	cggtagcggt	gatcgtgcag	gacgagggag	tggacggcat	4440
cccgggtgaag	gtcctcaact	gtgacaccat	ctcccaggtc	aaggagaaga	tcattgacca	4500
gggtatccgt	gggcagccct	gtcctctgtg	gccccaggca	gacagcgtgg	tcctggagtg	4560
gcgtccgggc	tccacagcgc	agatcctgtc	ggacctggac	ctgacgtcac	agcgggaggg	4620
ccggtgggaag	cgcgtcaaca	cccttatgca	ctacaatgtc	cgggatggag	ccacctcat	4680
cctgtccaag	gtgggggtct	ccagcagcc	ggaggacagc	cagcaggacc	tgccctggga	4740
gcgccatgcc	ctcctggagg	aggagaaccg	ggtgtggcac	ctggtgcggc	cgaccgacga	4800
gggtggacgag	ggcaagtcca	agagagggcag	cgtgaaagag	aaggagcgga	cgaaggccat	4860
caccgagatc	tacctgacgc	ggctgtcttc	agtcaagggc	acactgcagc	agttgtggga	4920
caacttcttc	cagagcgtgc	tggcgccctg	gcacgcgggt	ccacctgcag	tcaagtactt	4980
cttcgacttc	ctggacgagc	aggcagagaa	gcacaacatc	caggatgaag	acacactcca	5040

catctggaag	acgaacagct	taccgctccg	gttctgggtg	aacatcctca	agaaccccc	5100
cttcattctt	gacgtgcatg	tcacagaggt	ggtggagcgc	tcgctgtcag	tcattcgcgca	5160
gaccttcatg	gatgcctgca	cgcgcacgga	gcataagctg	agccgcgatt	ctccccagcaa	5220
caagctgtctg	tacgccaagg	agatctccac	ctacaagaag	atggtggagg	attactacaa	5280
ggggatccgg	cagatggtgc	aggtcagcga	ccaggacatg	aacacacacc	tggcagagat	5340
ttcccggggc	cacacgggact	ccttgaacac	cctcgtggca	ctccaccagc	tctaccaata	5400
cacgcagaag	tactatgacg	agatcatcaa	tgccttggag	gaggatcctg	ccgcccagaa	5460
gatgcagctg	gccttccgcc	tgacgagat	tgccgctgca	ctggagaaca	aggtcactga	5520
cctctgacct	acaatctcca	gtgctgcctt	gggacatagg	tacctgaggt	acctgagagc	5580
ccctcagggg	aggaggccga	gtggctgtgg	ctgaggcccc	cacctctccc	tggaaacgcg	5640
cccaagccgg	agtgggtgca	gccggaaccc	gccagcgctc	tagactgtag	catcttctct	5700
tgagcaatac	cgccggggcac	cgcaccagca	ccagccccag	ccccagctcc	ctccggccgc	5760
agaaccagca	tcgggtgttc	actgtcgagt	ctcgagtgat	ttgaaaatgt	gccttacgct	5820
gccacgctgg	gggcagctgg	cctccgcctc	cgccccagca	ccagcagccg	cctccatgcc	5880
ctagggttgg	cccctggggg	atctgagggc	ctgtggcccc	cagggcaagt	tcccagatcc	5940
tatgtctgtc	tgtccaccac	gagatgggag	gaggagaaaa	agcggtagca	tgccttctct	6000
acccctaccg	cctccccaa	gggtgcggca	ctctgggtgg	actcacggct	gctggggccc	6060
acgtcaaagg	tcaagtgaga	cgtaggtcaa	gtcctacgtc	ggggcccaga	catcctgggg	6120
tcctgggtctg	tcagacaggc	tgccttagag	ccccaccagg	tcggggggga	ctggggagcag	6180
ttccaagacc	accccacccc	tttttgtaaa	tcttgttcat	tgtaaatcaa	atacagcgct	6240
tttttcactc	cg					6252

<210> 2
 <211> 1838
 <212> PRT
 <213> HOMO SAPIEN

<400> 2
 Met Ala Leu Gln Leu Trp Ala Leu Thr Leu Leu Gly Leu Leu Gly Ala
 1 5 10 15
 Gly Ala Ser Leu Arg Pro Arg Lys Leu Asp Phe Phe Arg Ser Glu Lys
 20 25 30
 Glu Leu Asn His Leu Ala Val Asp Glu Ala Ser Gly Val Val Tyr Leu
 35 40 45
 Gly Ala Val Asn Ala Leu Tyr Gln Leu Asp Ala Lys Leu Gln Leu Glu
 50 55 60
 Gln Gln Val Ala Thr Gly Pro Ala Leu Asp Asn Lys Lys Cys Thr Pro
 65 70 75 80
 Pro Ile Glu Ala Ser Gln Cys His Glu Ala Glu Met Thr Asp Asn Val
 85 90 95
 Asn Gln Leu Leu Leu Asp Pro Pro Arg Lys Arg Leu Val Glu Cys
 100 105 110
 Gly Ser Leu Phe Lys Gly Ile Cys Ala Leu Arg Ala Leu Ser Asn Ile
 115 120 125
 Ser Leu Arg Leu Phe Tyr Glu Asp Gly Ser Gly Glu Lys Ser Phe Val
 130 135 140
 Ala Ser Asn Asp Glu Gly Val Ala Thr Val Gly Leu Val Ser Ser Thr
 145 150 155 160
 Gly Pro Gly Gly Asp Arg Val Leu Phe Val Gly Lys Gly Asn Gly Pro
 165 170 175
 His Asp Asn Gly Ile Ile Val Ser Thr Arg Leu Leu Asp Arg Thr Asp
 180 185 190
 Ser Arg Glu Ala Phe Glu Ala Tyr Thr Asp His Ala Thr Tyr Lys Ala

		195				200			205					
Gly	Tyr	Leu	Ser	Thr	Asn	Thr	Gln	Phe	Val	Ala	Ala	Phe	Glu	Asp
210						215				220				
Gly	Pro	Tyr	Val	Phe	Phe	Val	Phe	Asn	Gln	Gln	Asp	Lys	His	Pro
225					230					235				240
Arg	Asn	Arg	Thr	Leu	Leu	Ala	Arg	Met	Cys	Arg	Glu	Asp	Pro	Asn
					245				250				255	
Tyr	Ser	Tyr	Leu	Glu	Met	Asp	Leu	Gln	Cys	Arg	Asp	Pro	Asp	Ile
			260					265				270		His
Ala	Ala	Ala	Phe	Gly	Thr	Cys	Leu	Ala	Ala	Ser	Val	Ala	Ala	Pro
		275					280					285		Gly
Ser	Gly	Arg	Val	Leu	Tyr	Ala	Val	Phe	Ser	Arg	Asp	Ser	Arg	Ser
290						295					300			
Gly	Gly	Pro	Gly	Ala	Gly	Leu	Cys	Leu	Phe	Pro	Leu	Asp	Lys	Val
305					310					315				His
Ala	Lys	Met	Glu	Ala	Asn	Arg	Asn	Ala	Cys	Tyr	Thr	Gly	Thr	Arg
				325					330					Glu
Ala	Arg	Asp	Ile	Phe	Tyr	Lys	Pro	Phe	His	Gly	Asp	Ile	Gln	Cys
			340					345				350		Gly
Gly	His	Ala	Pro	Gly	Ser	Ser	Lys	Ser	Phe	Pro	Cys	Gly	Ser	Glu
		355					360					365		His
Leu	Pro	Tyr	Pro	Leu	Gly	Ser	Arg	Asp	Gly	Leu	Arg	Gly	Thr	Ala
370					375					380				Val
Leu	Gln	Arg	Gly	Gly	Leu	Asn	Leu	Thr	Ala	Val	Thr	Val	Ala	Ala
385					390					395				Glu
Asn	Asn	His	Thr	Val	Ala	Phe	Leu	Gly	Thr	Ser	Asp	Gly	Arg	Ile
			405					410						Leu
Lys	Val	Tyr	Leu	Thr	Pro	Asp	Gly	Thr	Ser	Ser	Glu	Tyr	Asp	Ser
			420					425					430	Ile
Leu	Val	Glu	Ile	Asn	Lys	Arg	Val	Lys	Arg	Asp	Leu	Val	Leu	Ser
		435					440					445		Gly
Asp	Leu	Gly	Ser	Leu	Tyr	Ala	Met	Thr	Gln	Asp	Lys	Val	Phe	Arg
450						455					460			Leu
Pro	Val	Gln	Glu	Cys	Leu	Ser	Tyr	Pro	Thr	Cys	Thr	Gln	Cys	Arg
465					470					475				Asp
Ser	Gln	Asp	Pro	Tyr	Cys	Gly	Trp	Cys	Val	Val	Glu	Gly	Arg	Cys
			485					490						Thr
Arg	Lys	Ala	Glu	Cys	Pro	Arg	Ala	Glu	Glu	Ala	Ser	His	Trp	Leu
		500						505					510	Trp
Ser	Arg	Ser	Lys	Ser	Cys	Val	Ala	Val	Thr	Ser	Ala	Gln	Pro	Gln
		515					520					525		Asn
Met	Ser	Arg	Arg	Ala	Gln	Gly	Glu	Val	Gln	Leu	Thr	Val	Ser	Pro
530						535					540			Leu
Pro	Ala	Leu	Ser	Glu	Glu	Asp	Glu	Leu	Leu	Cys	Leu	Phe	Gly	Glu
545					550					555				Ser
Pro	Pro	His	Pro	Ala	Arg	Val	Glu	Gly	Glu	Ala	Val	Ile	Cys	Asn
			565					570					575	Ser
Pro	Ser	Ser	Ile	Pro	Val	Thr	Pro	Pro	Gly	Gln	Asp	His	Val	Ala
		580						585					590	Val
Thr	Ile	Gln	Leu	Leu	Arg	Arg	Gly	Asn	Ile	Phe	Leu	Thr	Ser	Tyr
		595				600					605			
Gln	Tyr	Pro	Phe	Tyr	Asp	Cys	Arg	Gln	Ala	Met	Ser	Leu	Glu	Glu
610					615						620			Asn
Leu	Pro	Cys	Ile	Ser	Cys	Val	Ser	Asn	Arg	Trp	Thr	Cys	Gln	Trp
625					630					635				Asp

Leu Arg Tyr His Glu Cys Arg Glu Ala Ser Pro Asn Pro Glu Asp Gly
 645 650 655
 Ile Val Arg Ala His Met Glu Asp Ser Cys Pro Gln Phe Leu Gly Pro
 660 665 670
 Ser Pro Leu Val Ile Pro Met Asn His Glu Thr Asp Val Asn Phe Gln
 675 680 685
 Gly Lys Asn Leu Asp Thr Val Lys Gly Ser Ser Leu His Val Gly Ser
 690 700
 Asp Leu Leu Lys Phe Met Glu Pro Val Thr Met Gln Glu Ser Gly Thr
 705 710 715 720
 Phe Ala Phe Arg Thr Pro Lys Leu Ser His Asp Ala Asn Glu Thr Leu
 725 730 735
 Pro Leu His Leu Tyr Val Lys Ser Tyr Gly Lys Asn Ile Asp Ser Lys
 740 745 750
 Leu His Val Thr Leu Tyr Asn Cys Ser Phe Gly Arg Ser Asp Cys Ser
 755 760 765
 Leu Cys Arg Ala Ala Asn Pro Asp Tyr Arg Cys Ala Trp Cys Gly Gly
 770 775 780
 Gln Ser Arg Cys Val Tyr Glu Ala Leu Cys Asn Thr Thr Ser Glu Cys
 785 790 795 800
 Pro Pro Pro Val Ile Thr Arg Ile Gln Pro Glu Thr Gly Pro Leu Gly
 805 810 815
 Gly Gly Ile Arg Ile Thr Ile Leu Gly Ser Asn Leu Gly Val Gln Ala
 820 825 830
 Gly Asp Ile Gln Arg Ile Ser Val Ala Gly Arg Asn Cys Ser Phe Gln
 835 840 845
 Pro Glu Arg Tyr Ser Val Ser Thr Arg Ile Val Cys Val Ile Glu Ala
 850 855 860
 Ala Glu Thr Pro Phe Thr Gly Gly Val Glu Val Asp Val Phe Gly Lys
 865 870 875 880
 Leu Gly Arg Ser Pro Pro Asn Val Gln Phe Thr Phe Gln Gln Pro Lys
 885 890 895
 Pro Leu Ser Val Glu Pro Gln Gln Gly Gly Pro Gln Ala Gly Gly Thr Thr
 900 905 910
 Leu Thr Ile His Gly Thr His Leu Asp Thr Gly Ser Gln Glu Asp Val
 915 920 925
 Arg Val Thr Leu Asn Gly Val Pro Cys Lys Val Thr Lys Phe Gly Ala
 930 935 940
 Gln Leu Gln Cys Val Thr Gly Pro Gln Ala Thr Arg Gly Gln Met Leu
 945 950 955 960
 Leu Glu Val Ser Tyr Gly Gly Ser Pro Val Pro Asn Pro Gly Ile Phe
 965 970 975
 Phe Thr Tyr Arg Glu Asn Pro Val Leu Arg Ala Phe Glu Pro Leu Arg
 980 985 990
 Ser Phe Ala Ser Gly Gly Arg Ser Ile Asn Val Thr Gly Gln Gly Phe
 995 1000 1005
 Ser Leu Ile Gln Arg Phe Ala Met Val Val Ile Ala Glu Pro Leu Gln
 1010 1015 1020
 Ser Trp Gln Pro Pro Arg Glu Ala Glu Ser Leu Gln Pro Met Thr Val
 1025 1030 1035 1040
 Val Gly Thr Asp Tyr Val Phe His Asn Asp Thr Lys Val Val Phe Leu
 1045 1050 1055
 Ser Pro Ala Val Pro Glu Glu Pro Glu Ala Tyr Asn Leu Thr Val Leu
 1060 1065 1070
 Ile Glu Met Asp Gly His Arg Ala Leu Leu Arg Thr Glu Ala Gly Ala

1075				1080				1085							
Phe	Glu	Tyr	Val	Pro	Asp	Pro	Thr	Phe	Glu	Asn	Phe	Thr	Gly	Gly	Val
1090						1095					1100				
Lys	Lys	Gln	Val	Asn	Lys	Leu	Ile	His	Ala	Arg	Gly	Thr	Asn	Leu	Asn
1105				1110						1115				1120	
Lys	Ala	Met	Thr	Leu	Gln	Glu	Ala	Glu	Ala	Phe	Val	Gly	Ala	Glu	Arg
				1125						1130				1135	
Cys	Thr	Met	Lys	Thr	Leu	Thr	Glu	Thr	Asp	Leu	Tyr	Cys	Glu	Pro	Pro
				1140						1145				1150	
Glu	Val	Gln	Pro	Pro	Pro	Lys	Arg	Arg	Gln	Lys	Arg	Asp	Thr	Thr	His
				1155						1160				1165	
Asn	Leu	Pro	Glu	Phe	Ile	Val	Lys	Phe	Gly	Ser	Arg	Glu	Trp	Val	Leu
				1170						1175				1180	
Gly	Arg	Val	Glu	Tyr	Asp	Thr	Arg	Val	Ser	Asp	Val	Pro	Leu	Ser	Leu
1185				1190						1195				1200	
Ile	Leu	Pro	Leu	Val	Ile	Val	Pro	Met	Val	Val	Val	Ile	Ala	Val	Ser
				1205						1210				1215	
Val	Tyr	Cys	Tyr	Trp	Arg	Lys	Ser	Gln	Gln	Ala	Glu	Arg	Glu	Tyr	Glu
				1220						1225				1230	
Lys	Ile	Lys	Ser	Gln	Leu	Glu	Gly	Leu	Glu	Glu	Ser	Val	Arg	Asp	Arg
				1235						1240				1245	
Cys	Lys	Lys	Glu	Phe	Thr	Asp	Leu	Met	Ile	Glu	Met	Glu	Asp	Gln	Thr
				1250						1255				1260	
Asn	Asp	Val	His	Glu	Ala	Gly	Ile	Pro	Val	Leu	Asp	Tyr	Lys	Thr	Tyr
1265				1270						1275				1280	
Thr	Asp	Arg	Val	Phe	Phe	Leu	Pro	Ser	Lys	Asp	Gly	Asp	Lys	Asp	Val
				1285						1290				1295	
Met	Ile	Thr	Gly	Lys	Leu	Asp	Ile	Pro	Glu	Pro	Arg	Arg	Pro	Val	Val
				1300						1305				1310	
Glu	Gln	Ala	Leu	Tyr	Gln	Phe	Ser	Asn	Leu	Leu	Asn	Ser	Lys	Ser	Phe
				1315						1320				1325	
Leu	Ile	Asn	Phe	Ile	His	Thr	Leu	Glu	Asn	Gln	Arg	Glu	Phe	Ser	Ala
				1330						1335				1340	
Arg	Ala	Lys	Val	Tyr	Phe	Ala	Ser	Leu	Leu	Thr	Val	Ala	Leu	His	Gly
1345				1350						1355				1360	
Lys	Leu	Glu	Tyr	Tyr	Thr	Asp	Ile	Met	His	Thr	Leu	Phe	Leu	Glu	Leu
				1365						1370				1375	
Leu	Glu	Gln	Tyr	Val	Val	Ala	Lys	Asn	Pro	Lys	Leu	Met	Leu	Arg	Arg
				1380						1385				1390	
Ser	Glu	Thr	Val	Val	Glu	Arg	Met	Leu	Ser	Asn	Trp	Met	Ser	Ile	Cys
				1395						1400				1405	
Leu	Tyr	Gln	Tyr	Leu	Lys	Asp	Ser	Ala	Gly	Glu	Pro	Leu	Tyr	Lys	Leu
				1410						1415				1420	
Phe	Lys	Ala	Ile	Lys	His	Gln	Val	Glu	Lys	Gly	Pro	Val	Asp	Ala	Val
1425				1430						1435				1440	
Gln	Lys	Lys	Ala	Lys	Tyr	Thr	Leu	Asn	Asp	Thr	Gly	Leu	Leu	Gly	Asp
				1445						1450				1455	
Asp	Val	Glu	Tyr	Ala	Pro	Leu	Thr	Val	Ser	Val	Ile	Val	Gln	Asp	Glu
				1460						1465				1470	
Gly	Val	Asp	Ala	Ile	Pro	Val	Lys	Val	Leu	Asn	Cys	Asp	Thr	Ile	Ser
				1475						1480				1485	
Gln	Val	Lys	Glu	Lys	Ile	Ile	Asp	Gln	Val	Tyr	Arg	Gly	Gln	Pro	Cys
				1490						1495				1500	
Ser	Cys	Trp	Pro	Arg	Pro	Asp	Ser	Val	Val	Leu	Glu	Trp	Arg	Pro	Gly
1505				1510						1515				1520	

Ser Thr Ala Gln Ile Leu Ser Asp Leu Asp Leu Thr Ser Gln Arg Glu
 1525 1530 1535
 Gly Arg Trp Lys Arg Val Asn Thr Leu Met His Tyr Asn Val Arg Asp
 1540 1545 1550
 Gly Ala Thr Leu Ile Leu Ser Lys Val Gly Val Ser Gln Gln Pro Glu
 1555 1560 1565
 Asp Ser Gln Gln Asp Leu Pro Gly Glu Arg His Ala Leu Leu Glu Glu
 1570 1575 1580
 Glu Asn Arg Val Trp His Leu Val Arg Pro Thr Asp Glu Val Asp Glu
 1585 1590 1595 1600
 Gly Lys Ser Lys Arg Gly Ser Val Lys Glu Lys Glu Arg Thr Lys Ala
 1605 1610 1615
 Ile Thr Glu Ile Tyr Leu Thr Arg Leu Leu Ser Val Lys Gly Thr Leu
 1620 1625 1630
 Gln Gln Phe Val Asp Asn Phe Phe Gln Ser Val Leu Ala Pro Gly His
 1635 1640 1645
 Ala Val Pro Pro Ala Val Lys Tyr Phe Phe Asp Phe Leu Asp Glu Gln
 1650 1655 1660
 Ala Glu Lys His Asn Ile Gln Asp Glu Asp Thr Ile His Ile Trp Lys
 1665 1670 1675 1680
 Thr Asn Ser Leu Pro Leu Arg Phe Trp Val Asn Ile Leu Lys Asn Pro
 1685 1690 1695
 His Phe Ile Phe Asp Val His Val His Glu Val Val Asp Ala Ser Leu
 1700 1705 1710
 Ser Val Ile Ala Gln Thr Phe Met Asp Ala Cys Thr Arg Thr Glu His
 1715 1720 1725
 Lys Leu Ser Arg Asp Ser Pro Ser Asn Lys Leu Leu Tyr Ala Lys Glu
 1730 1735 1740
 Ile Ser Thr Tyr Lys Lys Met Val Glu Asp Tyr Tyr Lys Gly Ile Arg
 1745 1750 1755 1760
 Gln Met Val Gln Val Ser Asp Gln Asp Met Asn Thr His Leu Ala Glu
 1765 1770 1775
 Ile Ser Arg Ala His Thr Asp Ser Leu Asn Thr Leu Val Ala Leu His
 1780 1785 1790
 Gln Leu Tyr Gln Tyr Thr Gln Lys Tyr Tyr Asp Glu Ile Ile Asn Ala
 1795 1800 1805
 Leu Glu Glu Asp Pro Ala Ala Gln Lys Met Gln Leu Ala Phe Arg Leu
 1810 1815 1820
 Gln Gln Ile Ala Ala Ala Leu Glu Asn Lys Val Thr Asp Leu
 1825 1830 1835

<210> 3
 <211> 5367
 <212> DNA
 <213> HOMO SAPIEN

<400> 3
 atggctcgct ggccctccctt cggcctctgc ctccctcctgc tgctgctgtc cccaccgcca 60
 ctggcccttga cagggggccca tcgcttctcc gcaccttaata ccactctcaa ccacttggca 120
 ctggcacctg gccgagggcac actctatgtc ggcgcagtga accgcctctt ccagctcagc 180
 cccgagctgc agctcagggc cgtggctgtc actggccctg taatcgacag ccttgactgc 240
 gtgcccttcc gtgacccagc cgagtgcacca caggccccagc tcactgacaa tgccaaccag 300
 ctgctgctgg tgagcagccg cgcccaggag ctgggtggcct gcggggcaggt gcggcagggc 360
 gtgtgtgaga cacggcgccct tggggatgtg gccgaggtgc tgtaccaggc tgaggaccct 420
 ggtgacgggc agttttgtgc tgccaataacc ccgggagtggt caacgggtggg gctggtggtg 480

cccttgcgcc	gcccgggacct	cctgcttgtg	gccagaggcc	tggcggggcaa	gctgtcgcca	540
gggggtgccac	ccttggccat	ccgccagctg	gccgggtctc	agcccttctc	cagcgagggc	600
ctcggggccgc	tgggtgtggg	cgacttctcc	gactacaaca	acagctacgt	cggggccctt	660
gcgcagcccc	gtcccgccca	cttcgtgttc	cgccgcccgc	gggcccgggc	ccaggctgag	720
taccgtctct	acgtggcccg	cgctctgcctg	ggggacacca	acctgtactc	ctacgtggag	780
gtccccctcg	cccgccaggg	ccaggggcctc	atccaggccg	ccttctctgc	cccgggcacc	840
ttgctagggg	tgtttggccg	ggggcccaagg	ggcaccacagg	cggcgctctg	tgccctcccc	900
atggtggagc	tgggtgcccag	catggagcag	gccgggagac	tctgctacac	ggcggggcggc	960
cggggccccc	gcccgcgaga	ggaagccacc	gtggagtacg	gcgtcacgctc	gcgctcgctc	1020
accctgcccc	ttgatcccc	cgagtctgtac	ccctgtggcg	acgagcacac	ccccagcccc	1080
atttctggcc	gccagccctc	ggaggtccag	ccctgtctga	agctcgggca	gccggtcagc	1140
gccgtggcag	ctctccaggc	agatgggcac	atgatagcct	tcttggggga	caccaggggc	1200
cagctgtaca	agggtctttct	ccacggctcc	caggggccagg	tttaccactc	ccagcaagtg	1260
gggcctccag	gtccagccat	cagcccgagc	ctgctgctg	acagcagtgg	cagtcacctc	1320
tatgtccctg	ctgccaccac	ggtggaccgg	atacctgtgg	cagcctgccc	ccagtccctc	1380
gactgtgcca	gctgcctcca	ggcccaggac	ccgctgtgtg	gctggtgtgt	cctccagggc	1440
agggtaccc	ggaagggcca	gtgccccggg	gcaggccagc	tgaaccagtg	gctgtggagt	1500
tatgaggagg	acagccactg	cctgcacatc	cagagcctgc	tgcggggcca	ccacccccgc	1560
caggagcagg	gccaggtccac	tttgtctgtc	ccccggctgc	ccatctctga	tgcatagtaa	1620
tacttccatt	gtgcgttcgg	ggactatgac	agcttggctc	atgtggaagg	gccccacgtg	1680
gcctgtgtca	ccctccccca	agaccagggtg	ccacttaacc	ctccaggcac	agaccacgtc	1740
actgtgcccc	tggtccctgat	gttcgaggac	gtgactgtgg	ctgccaccaa	cttccctctt	1800
tatgactgca	gtgcccgtcca	ggccttggag	gcggctgccc	ccgtctcttc	ccaggggcctg	1860
cctgcctcct	tcactgtctg	gctggagctg	ctggagaac	ttcggggact	gcccggcacc	1920
ctggaggaga	cagcagggga	ttcaggccctc	atccactggc	agggccacca	gcggggagctc	1980
ccagtgcaca	tctactgtcc	ccagggtgaa	gcccagaggc	tggacaacac	ccatgctctt	2040
tatgtgtgagc	ctcaggcgagc	ccaggcaggc	ggggcagggtg	gggtggcaga	caggagcgcg	2100
tcagcacact	gtccagccctc	ccctagtgat	cctgtacgac	tgcccatatg	gccacccgga	2160
ctcagcccac	gcccaagcgg	cccaacaggag	ctggggctgc	ctgtgaccag	ccctccccca	2220
ggcccccaaa	ccccagcagc	tcggcctggc	tgggctggtt	ggctggccgg	gcaccagca	2280
ctgcagagtg	gagcgtgggt	gcggggggacc	ccatctgcca	tcatttgcct	gctgcaggtc	2340
gagccctctga	ccgttcccc	tgaggggaggc	ttggccctca	ccatctctgg	ctccaacctg	2400
ggccgggccc	tcgcccagtg	gcagtacgcc	gacctctgtc	tgctgagcct	gagtcctcgc	2460
tgggggcccc	aggcaggggg	caccagctc	accatccgag	gtcagcactc	ccagacaggt	2520
ggcaacacca	gtgccttctg	gggttgccaa	ccctgtccca	tgggtggggc	actgatccgt	2580
gtcaggggca	ccggcctaga	cgttgttcag	cgcccccctac	tgctctgtgtg	gctggaggct	2640
gacgcagagg	tgcaggcttc	caggggccag	ccccaggacc	cacagccaa	gaggagctgt	2700
ggagccctgt	ctgcggacc	ccaggcttgt	atccagctcg	gtggggggct	gctgcagcgc	2760
acagcagagc	ccagctcact	ccactgtgtg	tcggccctga	atgccccaca	gtgctccacc	2820
gtctgtctcg	tcaactctgc	cagcctcttc	ctgtgcggga	gcccctgtgt	accagacaga	2880
gcccaccgcg	agcgggtctt	cttcacccta	gacaacgtgc	aagtggactt	cgccagtgcc	2940
agtgggggcc	agggtctctc	gtaccagccc	aacccccgcc	tggcaccctc	cagccgcgag	3000
gggcctgccc	gcccctaccg	cctcaagcca	ggccatgtcc	tggatgtgga	gggcgagggc	3060
ctcaacctgt	gcctcagcaa	ggaggaggtg	cgctgtcaca	tcggcccgcg	cgagtgcctg	3120
gtgaagacgc	tcacgcgcac	ccacctgtac	tgcgagccgc	ctgcgcacgc	cccgagcctc	3180
gccaatggct	ccggcctgcc	acagtctctg	gtgcagatgg	gcaatgtgca	gctggccctg	3240
ggccctgtgc	agtacgaggc	tgaacccccg	ctgtctgcct	ttcccggtga	ggcccaggca	3300
ggcgtgggca	tgggtgtctg	agtgctgatt	gccgcccgtg	tcctctccac	cctcatgtac	3360
aggcacaaga	gcaagcaggc	cctgcgggac	taccagaagg	tgctagtgtca	gctggagagc	3420
ctggagagcg	gcgtgggaga	ccagtggccg	aaggagtcca	cagacctcat	gacggagatg	3480
accgacctca	cgagcgacct	ggaggcgagc	gggatccctc	tccttgacta	ccgcacctac	3540
gccgagcgcg	ccttcttccc	tggccatggc	ggttgcccgc	tgacgcccac	gcttgagggg	3600
ccagggggagg	acggccactg	tgccactgtg	cgccaggggc	tcacgcagct	ctccaacctg	3660
ctcaacagca	agctcttctc	cctcacgggtg	agggcgctgt	ggcgggagtg	cccagtgggc	3720
aaggagggtg	ggctggggaa	ctactgcctc	gagacaaagg	tggggggagg	gacagagacc	3780

atggtggaga	aactgctcac	caactggctg	tccatctgcc	tgtacgcctt	cctgagggag	3840
gtggctgggtg	aaccactgta	catgctcttc	cgggccatcc	agtaccaggt	ggacaaaggg	3900
cccgtggagc	ccgtgacagg	caaggccaaa	cggaccctga	atgatagccg	cttgctgcgg	3960
gaggacgtgg	agtccagcc	cctgacgctg	atggtgctgg	tggggcccgg	ggctggccgg	4020
gccgcaggca	gcagcgagat	gcagcgcgtg	ccagcccggg	tgctcgacac	ggacaccatc	4080
acccagggtca	aggagaaggt	gttggacca	gtctacaagg	gcaccccctt	ctcccagagg	4140
ccctcagtcg	atgccctaga	ccttggtgag	agagccagcc	ctgcccaccc	accccagggg	4200
cccttcacct	ccctccggc	acctggagcc	cctcaactgt	gtcttactat	gaacataccc	4260
acgctggagg	atggcgagg	gggggggggtg	tgctctggc	acctggtgaa	agccaccgag	4320
gagccagaag	gggccaaggt	gcggtgcagc	agcctgcggg	agcgcgagcc	agcaaggggc	4380
aaggccattc	cggaaatcta	cctcacccgt	ctgctgtcca	tgaagggtgg	tgccggcctgg	4440
gtggctgggc	ctgagaggag	gctcagccag	ggaccccagc	cgagccaggg	tgtgggaggg	4500
gcagggggcag	cctcagccgt	ggatggcccc	cacaccctgc	cctccacaca	gcccttatcc	4560
cctgcctcgc	agggcacgct	gcagaagttt	gtggacgaca	ccttccaggc	cattctcagc	4620
gtgaaccggc	ccatcccat	cgccgtcaag	tacctgtttg	accttctgga	tgagctagca	4680
gagaagcacg	gcacgcagga	cccaggggacc	ctgcacatct	ggaagaccac	cagtcctgctg	4740
ctgcgggtctt	gggtgaatgc	cttgaagaac	ccacagctca	tctttgatgt	acgggtgtcg	4800
gacaaatgtgg	acgccatcct	tgctgtcatc	gccagagcct	tcattgactc	ctgtaccacc	4860
tcggagcata	aagtggggccg	ggtgagagca	gtgccagcag	cagcagctgg	caggggcttg	4920
aggagggaag	gcttatgggg	gaagcctaga	gggctgtgca	cagagctctg	gtgtgggcagt	4980
ggcagcatca	tggggggcacc	ttcacctccg	agctcatgcc	tagccctccc	cctccctccg	5040
gagcaggatc	ccccagtgaa	caaacctgct	taagggccggg	agatccccag	ctacaagcag	5100
atggtggaga	ggtactatgc	ggacattcgc	cagagctctc	cggcgagcta	ccaggagatg	5160
aactctgctt	tggtctgagct	ctccgggaac	tacacttctg	ctccccactg	tctggaggct	5220
ctgcaagaac	tctacaacca	catccacagg	tactatgac	agattatcag	tgccctggag	5280
gaggaccctg	tgggccagaa	gctgcagctg	gcctgccggc	tgacagcaggt	cgccgcccctg	5340
gtggaaaaca	aagtgactga	cctgtga				5367

<210> 4
 <211> 1788
 <212> PRT
 <213> HOMO SAPIEN

<400> 4
 Met Ala Arg Trp Pro Pro Phe Gly Leu Cys Leu Leu Leu Leu Leu
 1 5 10 15
 Ser Pro Pro Pro Leu Pro Leu Thr Gly Ala His Arg Phe Ser Ala Pro
 20 25 30
 Asn Thr Thr Leu Asn His Leu Ala Leu Ala Pro Gly Arg Gly Thr Leu
 35 40 45
 Tyr Val Gly Ala Val Asn Arg Leu Phe Gln Leu Ser Pro Glu Leu Gln
 50 55 60
 Leu Glu Ala Val Ala Val Thr Gly Pro Val Ile Asp Ser Pro Asp Cys
 65 70 75 80
 Val Pro Phe Arg Asp Pro Ala Glu Cys Pro Gln Ala Gln Leu Thr Asp
 85 90 95
 Asn Ala Asn Gln Leu Leu Leu Val Ser Ser Arg Ala Gln Glu Leu Val
 100 105 110
 Ala Cys Gly Gln Val Arg Gln Gly Val Cys Glu Thr Arg Arg Leu Gly
 115 120 125
 Asp Val Ala Glu Val Leu Tyr Gln Ala Glu Asp Pro Gly Asp Gly Gln
 130 135 140
 Phe Val Ala Ala Asn Thr Pro Gly Val Ala Thr Val Gly Leu Val Val

```

145          150          155          160
Pro Leu Pro Gly Arg Asp Leu Leu Leu Val Ala Arg Gly Leu Ala Gly
Lys Leu Ser Ala Gly Val Pro Pro Leu Ala Ile Arg Gln Leu Ala Gly
Ser Gln Pro Phe Ser Ser Glu Gly Leu Gly Arg Leu Val Val Gly Asp
Phe Ser Asp Tyr Asn Asn Ser Tyr Val Gly Ala Phe Ala Asp Ala Arg
Ser Ala Tyr Phe Val Phe Arg Arg Arg Gly Ala Arg Ala Gln Ala Glu
Tyr Arg Ser Tyr Val Ala Arg Val Cys Leu Gly Asp Thr Asn Leu Tyr
Ser Tyr Val Glu Val Pro Leu Ala Cys Gln Gly Gln Gly Leu Ile Gln
Ala Ala Phe Leu Ala Pro Gly Thr Leu Leu Gly Val Phe Ala Ala Gly
Pro Arg Gly Thr Gln Ala Ala Leu Cys Ala Phe Pro Met Val Glu Leu
Gly Ala Ser Met Glu Gln Ala Arg Arg Leu Cys Tyr Thr Ala Gly Gly
Arg-Gly-Pro-Ser-Gly-Ala-Glu-Glu-Ala-Thr-Val-Glu-Tyr-Gly-Val-Thr
Ser Arg Cys Val Thr Leu Pro Leu Asp Ser Pro Glu Ser Tyr Pro Cys
Gly Asp Glu His Thr Pro Ser Pro Ile Ala Gly Arg Gln Pro Leu Glu
Val Gln Pro Leu Leu Lys Leu Gly Gln Pro Val Ser Ala Val Ala Ala
Leu Gln Ala Asp Gly His Met Ile Ala Phe Leu Gly Asp Thr Gln Gly
Gln Leu Tyr Lys Val Phe Leu His Gly Ser Gln Gly Gln Val Tyr His
Ser Gln Gln Val Gly Pro Pro Gly Ser Ala Ile Ser Pro Asp Leu Leu
Leu Asp Ser Ser Gly Ser His Leu Tyr Val Leu Thr Ala His Gln Val
Asp Arg Ile Pro Val Ala Ala Cys Pro Gln Phe Pro Asp Cys Ala Ser
Cys Leu Gln Ala Gln Asp Pro Leu Cys Gly Trp Cys Val Leu Gln Gly
Arg Cys Thr Arg Lys Gly Gln Cys Gly Arg Ala Gly Gln Leu Asn Gln
Trp Leu Trp Ser Tyr Glu Glu Asp Ser His Cys Leu His Ile Gln Ser
Leu Leu Pro Gly His His Pro Arg Gln Glu Gln Gly Gln Val Thr Leu
Ser Val Pro Arg Leu Pro Ile Leu Asp Ala Asp Glu Tyr Phe His Cys
Ala Phe Gly Asp Tyr Asp Ser Leu Ala His Val Glu Gly Pro His Val
Ala Cys Val Thr Pro Gln Asp Gln Val Pro Leu Asn Pro Pro Gly
Thr Asp His Val Thr Val Pro Leu Ala Leu Met Phe Glu Asp Val Thr
          580          585          590

```

Val Ala Ala Thr Asn Phe Ser Phe Tyr Asp Cys Ser Ala Val Gln Ala
 595 600 605
 Leu Glu Ala Ala Pro Val Leu Pro Gln Gly Leu Pro Ala Ser Phe
 610 615 620
 His Cys Trp Leu Glu Leu Pro Gly Glu Leu Arg Gly Leu Pro Ala Thr
 625 630 635 640
 Leu Glu Glu Thr Ala Gly Asp Ser Gly Leu Ile His Cys Gln Ala His
 645 650 655
 Gln Arg Glu Leu Pro Val Pro Ile Tyr Val Thr Gln Gly Glu Ala Gln
 660 665 670
 Arg Leu Asp Asn Thr His Ala Leu Tyr Gly Glu Pro Glu Gly Ser Gln
 675 680 685
 Ala Gly Gly Ala Gly Trp Val Ala Asp Arg Arg Arg Ser Ala His Cys
 690 695 700
 Leu Thr Leu Pro Ser Asp Pro Val Arg Leu Arg His Gly Pro Pro Gly
 705 710 715 720
 Leu Gln Pro Leu Pro Ser Gly Gln Gln Glu Pro Gly Leu Pro Val Thr
 725 730 735
 Ser Pro Ala Pro Gly Pro Gln Thr Pro Ala Ala Arg Pro Gly Trp Ala
 740 745 750
 Gly Trp Leu Ala Gly His Pro Ala Leu Gln Ser Gly Ala Trp Val Arg
 755 760 765
 Gly Thr Pro Ser Ala Ile Ile Cys Leu Leu Gln Val Gln Pro Leu Thr
 770 775 780
 Gly Pro Pro Glu Gly Gly Leu Ala Leu Thr Ile Leu Gly Ser Asn Leu
 785 790 795 800
 Gly Arg Ala Phe Ala Asp Val Gln Tyr Ala Asp Pro Val Leu Leu Ser
 805 810 815
 Leu Ser Pro Arg Trp Gly Pro Gln Ala Gly Gly Thr Gln Leu Thr Ile
 820 825 830
 Arg Gly Gln His Leu Gln Thr Gly Gly Asn Thr Ser Ala Phe Val Gly
 835 840 845
 Gly Gln Pro Cys Pro Met Gly Gly Arg Leu Ile Arg Val Arg Gly Thr
 850 855 860
 Gly Leu Asp Val Val Gln Arg Pro Leu Leu Ser Val Trp Leu Glu Ala
 865 870 875 880
 Asp Ala Glu Val Gln Ala Ser Arg Ala Gln Pro Gln Asp Pro Gln Pro
 885 890 895
 Arg Arg Ser Cys Gly Ala Pro Ala Ala Asp Pro Gln Ala Cys Ile Gln
 900 905 910
 Leu Gly Gly Gly Leu Leu Gln Arg Thr Ala Glu Pro Ser Ser Leu His
 915 920 925
 Leu Trp Ser Ala Leu Asn Ala Pro Gln Cys Ser Thr Val Cys Ser Val
 930 935 940
 Asn Ser Ser Ser Leu Leu Leu Cys Arg Ser Pro Ala Val Pro Asp Arg
 945 950 955 960
 Ala His Pro Gln Arg Val Phe Phe Thr Leu Asp Asn Val Gln Val Asp
 965 970 975
 Phe Ala Ser Ala Ser Gly Gly Gln Gly Phe Leu Tyr Gln Pro Asn Pro
 980 985 990
 Arg Leu Ala Pro Leu Ser Arg Glu Gly Pro Ala Arg Pro Tyr Arg Leu
 995 1000 1005
 Lys Pro Gly His Val Leu Asp Val Glu Gly Glu Gly Leu Asn Leu Gly
 1010 1015 1020
 Ile Ser Lys Glu Glu Val Arg Val His Ile Gly Arg Gly Glu Cys Leu

1025		1030		1035		1040
Val Lys Thr Leu Thr Arg Thr His Leu Tyr Cys Glu Pro Pro Ala His						
1045		1045		1050		1055
Ala Pro Gln Pro Ala Asn Gly Ser Gly Leu Pro Gln Phe Val Val Gln						
1060		1065		1070		
Met Gly Asn Val Gln Leu Ala Leu Gly Pro Val Gln Tyr Glu Ala Glu						
1075		1080		1085		
Pro Pro Leu Ser Ala Phe Pro Val Glu Ala Gln Ala Gly Val Gly Met						
1090		1095		1100		
Gly Ala Ala Val Leu Ile Ala Ala Val Leu Leu Thr Leu Met Tyr						
1105		1110		1115		1120
Arg His Lys Ser Lys Gln Ala Leu Arg Asp Tyr Gln Lys Val Leu Val						
1125		1130		1135		
Gln Leu Glu Ser Leu Glu Thr Gly Val Gly Asp Gln Cys Arg Lys Glu						
1140		1145		1150		
Phe Thr Asp Leu Met Thr Glu Met Thr Asp Leu Ser Ser Asp Leu Glu						
1155		1160		1165		
Gly Ser Gly Ile Pro Phe Leu Asp Tyr Arg Thr Tyr Ala Glu Arg Ala						
1170		1175		1180		
Phe Phe Pro Gly His Gly Gly Cys Pro Leu Gln Pro Lys Pro Glu Gly						
1185		1190		1195		1200
Pro Gly Glu Asp Gly His Cys Ala Thr Val Arg Gln Gly Leu Thr Gln						
1205		1210		1215		
Leu Ser Asn Leu Leu Asn Ser Lys Leu Phe Leu Leu Thr Val Arg Ala						
1220		1225		1230		
Val Trp Arg Glu Cys Pro Val Gly Lys Glu Val Gly Leu Gly Asn Tyr						
1235		1240		1245		
Trp Pro Glu Thr Lys Val Gly Glu Glu Thr Glu Thr Met Val Glu Lys						
1250		1255		1260		
Leu Leu Thr Asn Trp Leu Ser Ile Cys Leu Tyr Ala Phe Leu Arg Glu						
1265		1270		1275		1280
Val Ala Gly Glu Pro Leu Tyr Met Leu Phe Arg Ala Ile Gln Tyr Gln						
1285		1290		1295		
Val Asp Lys Gly Pro Val Asp Ala Val Thr Gly Lys Ala Lys Arg Thr						
1300		1305		1310		
Leu Asn Asp Ser Arg Leu Leu Arg Glu Asp Val Glu Phe Gln Pro Leu						
1315		1320		1325		
Thr Leu Met Val Leu Val Gly Pro Gly Ala Gly Gly Ala Ala Gly Ser						
1330		1335		1340		
Ser Glu Met Gln Arg Val Pro Ala Arg Val Leu Asp Thr Asp Thr Ile						
1345		1350		1355		1360
Thr Gln Val Lys Glu Lys Val Leu Asp Gln Val Tyr Lys Gly Thr Pro						
1365		1370		1375		
Phe Ser Gln Arg Pro Ser Val His Ala Leu Asp Leu Gly Glu Arg Ala						
1380		1385		1390		
Ser Pro Ala His Pro Pro Gln Gly Pro Phe Pro Thr Pro Pro Ala Pro						
1395		1400		1405		
Gly Ala Pro Gln Leu Cys Leu Thr Met Asn Ile Pro Thr Leu Glu Asp						
1410		1415		1420		
Gly Glu Glu Gly Gly Val Cys Leu Trp His Leu Val Lys Ala Thr Glu						
1425		1430		1435		1440
Glu Pro Glu Gly Ala Lys Val Arg Cys Ser Ser Leu Arg Glu Arg Glu						
1445		1450		1455		
Pro Ala Arg Ala Lys Ala Ile Pro Glu Ile Tyr Leu Thr Arg Leu Leu						
1460		1465		1470		

Ser Met Lys Val Gly Ala Ala Trp Val Ala Gly Pro Glu Arg Arg Leu
 1475 1480 1485
 Ser Gln Gly Pro Arg Pro Ser Gln Gly Val Gly Gly Ala Ala Ala
 1490 1495 1500
 Ser Ala Val Asp Gly Pro His Thr Leu Pro Ser Thr Gln Pro Leu Ser
 1505 1510 1515
 Pro Ala Ser Gln Gly Thr Leu Gln Lys Phe Val Asp Asp Thr Phe Gln
 1525 1530 1535
 Ala Ile Leu Ser Val Asn Arg Pro Ile Pro Ile Ala Val Lys Tyr Leu
 1540 1545 1550
 Phe Asp Leu Leu Asp Glu Leu Ala Glu Lys His Gly Ile Glu Asp Pro
 1555 1560 1565
 Gly Thr Leu His Ile Trp Lys Thr Asn Ser Leu Leu Leu Arg Phe Trp
 1570 1575 1580
 Val Asn Ala Leu Lys Asn Pro Gln Leu Ile Phe Asp Val Arg Val Ser
 1585 1590 1595
 Asp Asn Val Asp Ala Ile Leu Ala Val Ile Ala Gln Thr Phe Ile Asp
 1605 1610 1615
 Ser Cys Thr Thr Ser Glu His Lys Val Gly Arg Val Arg Ala Val Pro
 1620 1625 1630
 Ala Ala Ala Gly Arg Gly Leu Arg Arg Lys Gly Leu Trp Gly Lys
 1635 1640 1645
 Pro Arg Gly Leu Cys Thr Glu Leu Trp Val Gly Ser Gly Ser Ile Met
 1650 1655 1660
 Gly Ala Pro Ser Pro Pro Ser Ser Cys Leu Ala Pro Pro Leu Pro Pro
 1665 1670 1675
 Glu Gln Asp Ser Pro Val Asn Lys Leu Leu Tyr Ala Arg Glu Ile Pro
 1685 1690 1695
 Arg Tyr Lys Gln Met Val Glu Arg Tyr Tyr Ala Asp Ile Arg Gln Ser
 1700 1705 1710
 Ser Pro Ala Ser Tyr Gln Glu Met Asn Ser Ala Leu Ala Glu Leu Ser
 1715 1720 1725
 Gly Asn Tyr Thr Ser Ala Pro His Cys Leu Glu Ala Leu Gln Glu Leu
 1730 1735 1740
 Tyr Asn His Ile His Arg Tyr Tyr Asp Gln Ile Ile Ser Ala Leu Glu
 1745 1750 1755
 Glu Asp Pro Val Gly Gln Lys Leu Gln Leu Ala Cys Arg Leu Gln Gln
 1765 1770 1775
 Val Ala Ala Leu Val Glu Asn Lys Val Thr Asp Leu
 1780 1785

<210> 5
 <211> 5892
 <212> DNA
 <213> HOMO SAPIEN

<400> 5
 ggcacagccc ggatggctct tcgcgccgcg ggcggcgcac cctttagcgg ccgggccgcc 60
 gctgccagcc ccccgccgtt ccagacgccg ccgcggtgcc cggtgccgct gctgttgctg 120
 ctgctcctgg gggcgccgcy ggcggcgccc ctggagatcc agcgtcggtt cccctcgccc 180
 acgcccacca acaacttcgc cctggacggc ggcgggggga ccgtgtacct ggcggccgtc 240
 aaccgcctct atcagctgtc gggcgccaac ctgagcctgg aggcggaggg ggcctggggc 300
 ccggtgcccg acagcccgcgt gtgtcacgct ccgcagctgc gcgagggcct gtgcgagcac 360
 ccggcgccgc tcacggacaa ctacaacaag atcctgcagc tggaccccg ccagggcctg 420
 gtagtcgtgt gcgggtccat ctaccagggc ttctgccagc tgcggcgccc gggtaacatc 480

tcggccgctgt	ccgtgcgctt	cccgcgccgc	gcgcgcgccg	ccgagcccg	cacggtgttc	540
cccagcatgc	tgaacgtggc	ggccaaccac	ccgaaccggt	ccaccgtggg	gctagtctcg	600
ctctcccgcc	cggcgccggg	ggcgaccg	ctgctcgtgg	gcgccacgta	caccggttac	660
ggcagctcct	tcttcccgcg	caaccgcagc	ctggaggacc	accgcttcga	gaacacgccc	720
gagatcgcca	tccgctccct	ggacacgcgc	ggcgacctgg	ccaagctctt	caccttcgac	780
ctcaacccct	ccgacgacaa	catctctcaag	atcaagcagg	gcgccaagga	gcagcacaa	840
ctgggctctg	tgagcgccct	cctgcacccg	tccgacccgc	cgccgggtgc	acagtcctac	900
gcgtacctgc	cgtctcaacag	cgaggcgccg	gcggcgcgaca	aggagagcca	ggcgcggaagc	960
ctgctggcgc	gcactctgcct	gccccacggc	gcggcgccgc	acgccaagaa	gctcaccgag	1020
tccracatcc	agttggcgct	cgagtgcgcg	ggcgcgccgc	gcccgcgcga	cctctacagc	1080
cgcttggtgt	cggtcttccc	agcccgggag	cggtctcttg	ctgtcttcga	gcggcccccag	1140
gggtcccccg	cggcccgccg	tgctccggcc	gcactctcgc	cttctccgctt	cgccgacggtg	1200
cgagcgcgca	tccgagctgc	gcgcaccgcc	tgcttcgtgg	aaccggcgcc	cgactgggtg	1260
gcggtgctcg	acagcgttgt	gcagggcacg	ggaccggcct	gcgagcgcaa	gctcaacatc	1320
cagctccagc	cagagcagct	ggacttgtga	gctgctcacc	tgacgacccc	gctgtccatc	1380
ctcgagcccc	tgaaggccac	gccccgtgtc	cgcccccggg	gcctcacctc	cgctggccgtg	1440
gcccagctca	acaactacac	agcgggtcttc	ctgggcacgg	tcaacgggag	gcttctcaag	1500
atcaacctga	acgagagcat	gcagtggtgt	agcaggcggg	tgggtgactgt	ggcctatggg	1560
gagcccgctg	accatgtcat	gcagtttgac	ccagcagact	ccggttacct	ttacctgatg	1620
acgtccccc	agatggccag	ggatgaagtc	gcgcctgca	acgtgcactc	cactctgtggg	1680
gactcgctgg	gtgcggcgga	cgccctactgc	ggctggtgtg	ccctggagac	gcggtgcacc	1740
ttcgacagag	actgcaccaa	tcccagccag	cagcatcttc	ggaccagtgc	cagcgagggc	1800
ccagcccgct	gtcttgcctt	gaccgtctctg	cttctccgaga	tcgagtggcg	ccaggagtac	1860
ccaggcctga	tcttgacagt	ctcgccagcc	ctcagtgcat	gcagatggcc	gcagatggcc	1920
tgtgactatg	ggaacaacat	ccgcactgtg	gctcgggtcc	caggccctgc	ctttggtcac	1980
cagattgctc	actgcaacct	cctgccgagg	gaccagtctc	cgcccttccc	ccccaaaccag	2040
gaccacgtga	gctgttagat	gtcaatgggc	ggaacatcgt	caaggccaat	caaggccaat	2100
ttaccatctc	acgactgcag	ccgcactgca	caagtgtacc	cccacacagc	ctgtaccagc	2160
tgctcttcgg	cacagtggcc	ctgtttcttg	tgacgcccag	agcactcctg	tgtttccaac	2220
cagtctcgtg	gcgaggccct	accaaaccct	acgagccctc	aggactgccc	ccggaccctg	2280
ctctcaccct	tggcaccggt	gcctaccggt	ggctcccaga	acatcctggt	gcctctggcc	2340
aacactgcct	ttttccaggg	tgacgcccctg	gagtgtagtt	ttgggctgga	ggagatcttc	2400
gaggctgtgt	gggtgaaatg	gtctgttgta	cgctgtgacc	aggtggtgct	gcacacgacc	2460
cggaaagacc	aggtgttccc	gctcagccct	caactaaagg	ggcgggcgct	ccgatctctg	2520
gacagccctg	agcccatgac	agtcactggtc	tataactgtg	ccatgggcag	ccccgactgt	2580
tcccagtgcc	tgggcccgcg	agacctgggt	cacctgtgcg	tgtggagtga	ggctcctggc	2640
ctcgccgggg	ctctgcagcc	catggctggc	acctgcccgc	cccccgagat	ccgcgcgatt	2700
gagccctctga	gtggcccggt	ggacgggtgg	acctgtctga	ccatccgagg	aaggaaacctg	2760
ggccggcgccg	tcagtgacgt	ggcccacggc	gtgtggattg	gtggtgtggc	ctgtgagcca	2820
gtccctgaca	gatacacggt	gtcggaggag	atcgtgtgtg	tcacagggcc	agccccagga	2880
ccgtctctag	gtgtggtgac	cgtgaacgcc	tctaaggagg	gcaagtcccg	ggaccgcttc	2940
tccctacgtg	tgccccctgt	ccactcccctg	gagccctacca	tgggcccctaa	ggccggggggc	3000
accaggatca	ccatccatgg	gaatgacctc	catgtaggct	ccgagctcca	ggctcctgggt	3060
aacgacacag	accctctgac	ggagctgatg	cgacacagata	ccagcatcgc	ctgcaccatg	3120
cccgaggggg	ctccgcccgc	tccggtgcct	gtgtgtgtgc	gcttcgagcg	tccgggctgc	3180
gtgcacggga	acctcacctt	ctgggtacatg	cagaacccgg	tcatcacggc	catcagctcc	3240
cgccgcagcc	ctgtcagtg	cgccaggacc	atcacagtgg	ctggtgagcg	tttccacatg	3300
gtccagaatg	tgtccatggc	cgtccaccac	attggccggg	agccccagct	ctgcaagggt	3360
ctcaactcca	ccctcatcac	ctgcccgtcc	cccgggggccc	tgagcaacgc	atcagcgcca	3420
gtggacttct	tcactcaatg	gcgggcccac	gcagacgagg	tggctgtggc	tgaggagcta	3480
ctggaccccc	aggaggcaca	ggggggcagc	aggttccgccc	tggactacct	ccccaaacca	3540
cagttctctc	cggccaagag	ggagaaagtg	atcaagcacc	accccgggga	gcctctcacc	3600
ctcgttatcc	acaaggagag	ggacagcctg	gggctccaga	gtcacgagta	ccgggtcaag	3660
ataggccaag	taagctcgca	catccagatt	gtctctgaca	gaatcatcca	ctgctcggtc	3720
aacgagctcc	tgggcgcggc	cgtggggcag	ctgcccatac	caatccaggt	agggaacttc	3780


```

aaccagacca tcgccacact gcagctgggg ggcagcgaga cgcccatcat cgtgtccatc 3840
gtcatctgca gcgtcctgct gctgctctcc gtggtggccc tgttcgtctt ctgtaccaag 3900
agccgacgtg ctgagcggtta ctggcagaag acgctgctgc agatggagga gatggaatct 3960
cagatccgag aggaatccg caaaggcttc gctgagctgc agacagacat gacagatctg 4020
accaaggagc tgaaccgcag ccaggggcatt cctctcctgg agtataagca ctccgtgacc 4080
cgacacctct tcccgaagt ttctctcctt tatgaagagc gttacgtgct gccctcccag 4140
accctcaact cccaggggag ctcccaggga caggaaaccc acccacttgt gggagagtgg 4200
aagattcctg agagctgcg gcccaacatg gaagagggaa ttacgtgttt ctctcacta 4260
ctcaacaaca agcacttctt catcgtcttt gtccacgcgc tggagcagca gaaggacttt 4320
gcggtgcgcy acaggtgcag cctggcctcg ctgctgacca tcgcgtgca cggcaagctg 4380
gagtactaca ccagcatcat gaaggagctg ctggtggacc tcattgacgc ctccggccgc 4440
aagaaccccc agctcatgct gcggcgacac gagtctgtgg tggagaagat gctcaccaac 4500
tggatgtcca tctgcatgta cagctgtctg cgggagacgg tgggggagcc attcttctg 4560
ctgctgtgtg ccatcaagca gcaaatcaac aagggtctca tcgacgccat cacaggcaag 4620
gcccgtcata caatcaatga ggagtggctg ctgcgggaga acatcgagcc caagcccgg 4680
aaactgaacg tgccttcca gggctgtggc atggactcgc tgagcgtgcy ggcattggac 4740
accgacacgc tgacacaggt caaggagaag atcctggagg ccttctgcaa gaatgtgccc 4800
tactccaggt ggccgcgtgc agaggacgtc gaccttgagt ggttcgcctc cagcacacag 4860
agctacatcc ttccgggacct ggacgacacc tcagtgtggt aagacggccg caagaagctt 4920
aacacgctgg ccattacaa gatccctgaa ggtgcctccc tggccatgag tctcatagac 4980
aagaaggaca acacactggg ccgagtgaag gacttggaca cagagaagta ttccatttg 5040
-gtgcgtccta cggacgagct ggcggagccc aagaaagtct accggcagag ccactcgcaag 5100
aaggtgtctc cggaaatcta cctgaccgcg ctgctctcca ccaaggggac gttgcagaag 5160
ttcttgatg acctgttcaa ggcattctg agtatccgtg aagacaagcc cccactggct 5220
gtcaagtact ttctcgactt cctggaggag caggctgaga agaggggaat ctccgacccc 5280
gacaccttac acatctggaa gaccaacagc ctctctctcc ggttctgggt gaacatctg 5340
aagaaccccc agtttgtctt tgacatcgac aagacagacc acatcgagcc ctgcctttca 5400
gtcatcgccg aggccttcat cgacgcctgc tccatctctg acctgcagct gggcaaggat 5460
tcgccaacca acaagctcct ctacgccaag gagattcctg agtaccggaa gatcgtgcag 5520
cgctactaca agcagatcca ggacatgacg ccgctcagcg agcaagagat gaatgcccat 5580
ctggccgagg agtcgaggaa ataccagaat gagttcaaca ccaatgtggc catggcagag 5640
atttttaggt cgcccaagag gtatcgccg cagatcatgg ccgcgtgga ggccaacccc 5700
acggcccgga ggacacaact gcagcacaa tttgagcagg tgggtggctt gatggaggac 5760
aacatctacg agtgctacag tgaggcctga gacacatgga gagttggcta ggctgctgct 5820
gggagaaatg gacgcccact gggcctcaac ttgatctctt accccgtgcc tgtgactcag 5880
actgggaaat ac

```

<210> 6

<211> 1925

<212> PRT

<213> HOMO SAPIEN

<400> 6

```

Met Ala Leu Arg Ala Ala Gly Gly Ala Pro Phe Ser Gly Pro Ala Ala
1      5      10      15
Ala Ala Ser Pro Pro Phe Gln Thr Pro Arg Cys Pro Val Pro
20      25      30
Leu Leu Leu Leu Leu Leu Gly Ala Ala Arg Ala Gly Ala Leu Glu
35      40      45
Ile Gln Arg Arg Phe Pro Ser Pro Thr Pro Thr Asn Asn Phe Ala Leu
50      55      60
Asp Gly Ala Ala Gly Thr Val Tyr Leu Ala Ala Val Asn Arg Leu Tyr
65      70      75      80

```

Gln Leu Ser Gly Ala Asn Leu Ser Leu Glu Ala Glu Ala Ala Val Gly
 85 90 95
 Pro Val Pro Asp Ser Pro Leu Cys His Ala Pro Gln Leu Pro Gln Ala
 100 105 110
 Ser Cys Glu His Pro Arg Arg Leu Thr Asp Asn Tyr Asn Lys Ile Leu
 115 120 125
 Gln Leu Asp Pro Gly Gln Gly Leu Val Val Val Cys Gly Ser Ile Tyr
 130 135 140
 Gln Gly Phe Cys Gln Leu Arg Arg Arg Gly Asn Ile Ser Ala Val Ala
 145 150 155 160
 Val Arg Phe Pro Pro Ala Ala Pro Pro Ala Glu Pro Val Thr Val Phe
 165 170 175
 Pro Ser Met Leu Asn Val Ala Ala Asn His Pro Asn Ala Ser Thr Val
 180 185 190
 Gly Leu Val Leu Pro Pro Ala Ala Gly Ala Gly Gly Ser Arg Leu Leu
 195 200 205
 Val Gly Ala Thr Tyr Thr Gly Tyr Gly Ser Ser Phe Phe Pro Arg Asn
 210 215 220
 Arg Ser Leu Glu Asp His Arg Phe Glu Asn Thr Pro Glu Ile Ala Ile
 225 230 235 240
 Arg Ser Leu Asp Thr Arg Gly Asp Leu Ala Lys Leu Phe Thr Phe Asp
 245 250 255
 Leu Asn Pro Ser Asp Asp Asn Ile Leu Lys Ile Lys Gln Gly Ala Lys
 260 265 270
 Glu Gln His Lys Leu Gly Phe Val Ser Ala Phe Leu His Pro Ser Asp
 275 280 285
 Pro Pro Pro Gly Ala Gln Ser Tyr Ala Tyr Leu Ala Leu Asn Ser Glu
 290 295 300
 Ala Arg Ala Gly Asp Lys Glu Ser Gln Ala Arg Ser Leu Leu Ala Arg
 305 310 315 320
 Ile Cys Leu Pro His Gly Ala Gly Gly Asp Ala Lys Lys Leu Thr Glu
 325 330 335
 Ser Tyr Ile Gln Leu Gly Leu Gln Cys Ala Gly Gly Ala Gly Arg Gly
 340 345 350
 Asp Leu Tyr Ser Arg Leu Val Ser Val Phe Pro Ala Arg Glu Arg Leu
 355 360 365
 Phe Ala Val Phe Glu Arg Pro Gln Gly Ser Pro Ala Ala Arg Ala Ala
 370 375 380
 Pro Ala Ala Leu Cys Ala Phe Arg Phe Ala Asp Val Arg Ala Ala Ile
 385 390 395 400
 Arg Ala Ala Arg Thr Ala Cys Phe Val Glu Pro Ala Pro Asp Val Val
 405 410 415
 Ala Val Leu Asp Ser Val Val Gln Gly Thr Gly Pro Ala Cys Glu Arg
 420 425 430
 Lys Leu Asn Ile Gln Leu Gln Pro Glu Gln Leu Asp Cys Gly Ala Ala
 435 440 445
 His Leu Gln His Pro Leu Ser Ile Leu Gln Pro Leu Lys Ala Thr Pro
 450 455 460
 Val Phe Arg Ala Pro Gly Leu Thr Ser Val Ala Val Ala Ser Val Asn
 465 470 475 480
 Asn Tyr Thr Ala Val Phe Leu Gly Thr Val Asn Gly Arg Leu Leu Lys
 485 490 495
 Ile Asn Leu Asn Glu Ser Met Gln Val Val Ser Arg Arg Val Val Thr
 500 505 510
 Val Ala Tyr Gly Glu Pro Val His His Val Met Gln Phe Asp Pro Ala

515	520	525
Asp Ser Gly Tyr Leu Tyr Leu Met Thr Ser His Gln Met Ala Arg Val		
530	535	540
Lys Val Ala Ala Cys Asn Val His Ser Thr Cys Gly Asp Cys Val Gly		
545	550	555
Ala Ala Asp Ala Tyr Cys Gly Trp Cys Ala Leu Glu Thr Arg Cys Thr		
565	570	575
Leu Gln Gln Asp Cys Thr Asn Ser Ser Gln Gln His Phe Trp Thr Ser		
580	585	590
Ala Ser Glu Gly Pro Ser Arg Cys Pro Ala Met Thr Val Leu Pro Ser		
595	600	605
Glu Ile Asp Val Arg Gln Glu Tyr Pro Gly Met Ile Leu Gln Ile Ser		
610	615	620
Gly Ser Leu Pro Ser Leu Ser Gly Met Glu Met Ala Cys Asp Tyr Gly		
625	630	635
Asn Asn Ile Arg Thr Val Ala Arg Val Pro Gly Pro Ala Phe Gly His		
645	650	655
Gln Ile Ala Tyr Cys Asn Leu Leu Pro Arg Asp Gln Phe Pro Phe		
660	665	670
Pro Pro Asn Gln Asp His Val Thr Val Glu Met Ser Val Arg Val Asn		
675	680	685
Gly Arg Asn Ile Val Lys Ala Asn Phe Thr Ile Tyr Asp Cys Ser Arg		
690	695	700
Thr Ala Gln Val Tyr Pro His Thr Ala Cys Thr Ser Cys Leu Ser Ala		
705	710	715
Gln Trp Pro Cys Phe Trp Cys Ser Gln Gln His Ser Cys Val Ser Asn		
725	730	735
Gln Ser Arg Cys Glu Ala Ser Pro Asn Pro Thr Ser Pro Gln Asp Cys		
740	745	750
Pro Arg Thr Leu Leu Ser Pro Leu Ala Pro Val Pro Thr Gly Gly Ser		
755	760	765
Gln Asn Ile Leu Val Pro Leu Ala Asn Thr Ala Phe Phe Gln Gly Ala		
770	775	780
Ala Leu Glu Cys Ser Phe Gly Leu Glu Glu Ile Phe Glu Ala Val Trp		
785	790	795
Val Asn Glu Ser Val Arg Cys Asp Gln Val Val Leu His Thr Thr		
805	810	815
Arg Lys Ser Gln Val Phe Pro Leu Ser Leu Gln Leu Lys Gly Arg Pro		
820	825	830
Ala Arg Phe Leu Asp Ser Pro Glu Pro Met Thr Val Met Val Tyr Asn		
835	840	845
Cys Ala Met Gly Ser Pro Asp Cys Ser Gln Cys Leu Gly Arg Glu Asp		
850	855	860
Leu Gly His Leu Cys Val Trp Ser Asp Gly Cys Arg Leu Arg Gly Pro		
865	870	875
Leu Gln Pro Met Ala Gly Thr Cys Pro Ala Pro Glu Ile Arg Ala Ile		
885	890	895
Glu Pro Leu Ser Gly Pro Leu Asp Gly Gly Thr Leu Leu Thr Ile Arg		
900	905	910
Gly Arg Asn Leu Gly Arg Arg Leu Ser Asp Val Ala His Gly Val Trp		
915	920	925
Ile Gly Gly Val Ala Cys Glu Pro Leu Pro Asp Arg Tyr Thr Val Ser		
930	935	940
Glu Glu Ile Val Cys Val Thr Gly Pro Ala Pro Gly Pro Leu Ser Gly		
945	950	955

Val Val Thr Val Asn Ala Ser Lys Glu Gly Lys Ser Arg Asp Arg Phe
 965 970 975
 Ser Tyr Val Leu Pro Leu Val His Ser Leu Glu Pro Thr Met Gly Pro
 980 985 990
 Lys Ala Gly Gly Thr Arg Ile Thr Ile His Gly Asn Asp Leu His Val
 995 1000 1005
 Gly Ser Glu Leu Gln Val Leu Val Asn Asp Thr Asp Pro Cys Thr Glu
 1010 1015 1020
 Leu Met Arg Thr Asp Thr Ser Ile Ala Cys Thr Met Pro Glu Gly Ala
 1025 1030 1035
 Leu Pro Ala Pro Val Pro Val Cys Val Arg Phe Glu Arg Arg Gly Cys
 1045 1050 1055
 Val His Gly Asn Leu Thr Phe Trp Tyr Met Gln Asn Pro Val Ile Thr
 1060 1065 1070
 Ala Ile Ser Pro Arg Arg Ser Pro Val Ser Gly Gly Arg Thr Ile Thr
 1075 1080 1085
 Val Ala Gly Glu Arg Phe His Met Val Gln Asn Val Ser Met Ala Val
 1090 1095 1100
 His His Ile Gly Arg Glu Pro Thr Leu Cys Lys Val Leu Asn Ser Thr
 1105 1110 1115
 Leu Ile Thr Cys Pro Ser Pro Gly Ala Leu Ser Asn Ala Ser Ala Pro
 1125 1130 1135
 Val Asp Phe Phe Ile Asn Gly Arg Ala Tyr Ala Asp Glu Val Ala Val
 1140 1145 1150
 Ala Glu Glu Leu Leu Asp Pro Glu Glu Ala Gln Arg Gly Ser Arg Phe
 1155 1160 1165
 Arg Leu Asp Tyr Leu Pro Asn Pro Gln Phe Ser Thr Ala Lys Arg Glu
 1170 1175 1180
 Lys Trp Ile Lys His His Pro Gly Glu Pro Leu Thr Leu Val Ile His
 1185 1190 1195 1200
 Lys Glu Gln Asp Ser Leu Gly Leu Gln Ser His Glu Tyr Arg Val Lys
 1205 1210 1215
 Ile Gly Gln Val Ser Cys Asp Ile Gln Ile Val Ser Asp Arg Ile Ile
 1220 1225 1230
 His Cys Ser Val Asn Glu Ser Leu Gly Ala Ala Val Gly Gln Leu Pro
 1235 1240 1245
 Ile Thr Ile Gln Val Gly Asn Phe Asn Gln Thr Ile Ala Thr Leu Gln
 1250 1255 1260
 Leu Gly Gly Ser Glu Thr Ala Ile Ile Val Ser Ile Val Ile Cys Ser
 1265 1270 1275 1280
 Val Leu Leu Leu Leu Ser Val Val Ala Leu Phe Val Phe Cys Thr Lys
 1285 1290 1295
 Ser Arg Arg Ala Glu Arg Tyr Trp Gln Lys Thr Leu Leu Gln Met Glu
 1300 1305 1310
 Glu Met Glu Ser Gln Ile Arg Glu Glu Ile Arg Lys Gly Phe Ala Glu
 1315 1320 1325
 Leu Gln Thr Asp Met Thr Asp Leu Thr Lys Glu Leu Asn Arg Ser Gln
 1330 1335 1340
 Gly Ile Pro Phe Leu Glu Tyr Lys His Phe Val Thr Arg Thr Phe Phe
 1345 1350 1355 1360
 Pro Lys Cys Ser Ser Leu Tyr Glu Glu Arg Tyr Val Leu Pro Ser Gln
 1365 1370 1375
 Thr Leu Asn Ser Gln Gly Ser Ser Gln Ala Gln Glu Thr His Pro Leu
 1380 1385 1390
 Leu Gly Glu Trp Lys Ile Pro Glu Ser Cys Arg Pro Asn Met Glu Glu

1395	1400	1405
Gly Ile Ser Val Phe Ser Ser Leu Leu Asn Asn Lys His Phe Leu Ile		
1410	1415	1420
Val Phe Val His Ala Leu Glu Gln Gln Lys Asp Phe Ala Val Arg Asp		
1425	1430	1435
Arg Cys Ser Leu Ala Ser Leu Leu Thr Ile Ala Leu His Gly Lys Leu		1440
	1445	1450
Glu Tyr Tyr Thr Ser Ile Met Lys Glu Leu Leu Val Asp Leu Ile Asp		1455
	1460	1465
Ala Ser Ala Ala Lys Asn Pro Lys Leu Met Leu Arg Arg Thr Glu Ser		1470
	1475	1480
Val Val Glu Lys Met Leu Thr Asn Trp Met Ser Ile Cys Met Tyr Ser		1485
	1490	1495
Cys Leu Arg Glu Thr Val Gly Glu Pro Phe Phe Leu Leu Leu Cys Ala		1500
1505	1510	1515
Ile Lys Gln Gln Ile Asn Lys Gly Ser Ile Asp Ala Ile Thr Gly Lys		1520
	1525	1530
Ala Arg Tyr Thr Leu Asn Glu Glu Trp Leu Leu Arg Glu Asn Ile Glu		1535
	1540	1545
Ala Lys Pro Arg Asn Leu Asn Val Ser Phe Gln Gly Cys Gly Met Asp		1550
	1555	1560
Ser Leu Ser Val Arg Ala Met Asp Thr Asp Thr Leu Thr Gln Val Lys		1565
1570	1575	1580
Glu Lys Ile Leu Glu Ala Phe Cys Lys Asn Val Pro Tyr Ser Gln Trp		1585
	1590	1595
Pro Arg Ala Glu Asp Val Asp Leu Glu Trp Phe Ala Ser Ser Thr Gln		1600
	1605	1610
Ser Tyr Ile Leu Arg Asp Leu Asp Asp Thr Ser Val Val Glu Asp Gly		1615
	1620	1625
Arg Lys Lys Leu Asn Thr Leu Ala His Tyr Lys Ile Pro Glu Gly Ala		1630
	1635	1640
Ser Leu Ala Met Ser Leu Ile Asp Lys Lys Asp Asn Thr Leu Gly Arg		1645
	1650	1655
Val Lys Asp Leu Asp Thr Glu Lys Tyr Phe His Leu Val Leu Pro Thr		1660
	1665	1670
Asp Glu Leu Ala Glu Pro Lys Lys Ser His Arg Gln Ser His Arg Lys		1675
	1685	1690
Lys Val Leu Pro Glu Ile Tyr Leu Thr Arg Leu Leu Ser Thr Lys Gly		1695
	1700	1705
Thr Leu Gln Lys Phe Leu Asp Asp Leu Phe Lys Ala Ile Leu Ser Ile		1710
	1715	1720
Arg Glu Asp Lys Pro Pro Leu Ala Val Lys Tyr Phe Phe Asp Phe Leu		1725
	1730	1735
Glu Glu Gln Ala Glu Lys Arg Gly Ile Ser Asp Pro Asp Thr Leu His		1740
	1745	1750
Ile Trp Lys Thr Asn Ser Leu Pro Leu Arg Phe Trp Val Asn Ile Leu		1755
	1765	1770
Lys Asn Pro Gln Phe Val Phe Asp Ile Asp Lys Thr Asp His Ile Asp		1775
	1780	1785
Ala Cys Leu Ser Val Ile Ala Gln Ala Phe Ile Asp Ala Cys Ser Ile		1790
	1795	1800
Ser Asp Leu Gln Leu Gly Lys Asp Ser Pro Thr Asn Lys Leu Leu Tyr		1805
	1810	1815
Ala Lys Glu Ile Pro Glu Tyr Arg Lys Ile Val Gln Arg Tyr Tyr Lys		1820
1825	1830	1835
		1840

Gln Ile Gln Asp Met Thr Pro Leu Ser Glu Gln Glu Met Asn Ala His
 1845 1850 1855
 Leu Ala Glu Glu Ser Arg Lys Tyr Gln Asn Glu Phe Asn Thr Asn Val
 1860 1865 1870
 Ala Met Ala Glu Ile Phe Arg Ser Pro Lys Arg Tyr Arg Pro Gln Ile
 1875 1880 1885
 Met Ala Ala Leu Glu Ala Asn Pro Thr Ala Arg Arg Thr Gln Leu Gln
 1890 1895 1900
 His Lys Phe Glu Gln Val Val Ala Leu Met Glu Asp Asn Ile Tyr Glu
 1905 1910 1915 1920
 Cys Tyr Ser Glu Ala
 1925

<210> 7
 <211> 601
 <212> DNA
 <213> HOMO SAPIEN

<400> 7
 caccagagtc cctgtggagt cctgtgggtca gtatcagagc tgcggcgagt gccttgggtc 60
 agggcagccc cactgtggct ggtgtgtgtc gcacaacact tgcacccgga agggagcggg 120
 tgaagcgggtcc-aaggagcccc-gcaggtttgc-ctcggagatg_aagcagtggtg tccgggtgac 180
 ggtccatccc aacaatatct ccgtctctca gtacaacgcg ctgctgggtcc tggagacgta 240
 caatgtcccc gagctgtcag ctggcggtcaa ctgcaccttt gaggacctgt cagagatgga 300
 tgggctgggtc gtgggcaatc agatccagtg ctactcccct gcagccaagg aggtgccccg 360
 gatcatcaca gagaatgggg accaccatgt cgtacagctt cagctcaaat caaaggagac 420
 cggcatgacc ttcgccagca ccagctttgt cttctacaat tgcagcgtcc acaattcgtg 480
 cctgtcctgc gtggagagtc cataccgtg ccaactggtg aaataccggc atgtctgcac 540
 ccatgacccc aagacctgct ccttcaggga aggccgagt aagctgcccc aggttaggtcc 600
 c 601

<210> 8
 <211> 199
 <212> PRT
 <213> HOMO SAPIEN

<400> 8
 Thr Arg Val Pro Val Glu Ser Cys Gly Gln Tyr Gln Ser Cys Gly Glu
 1 5 10 15
 Cys Leu Gly Ser Gly Asp Pro His Cys Gly Trp Cys Val Leu His Asn
 20 25 30
 Thr Cys Thr Arg Lys Glu Arg Cys Glu Arg Ser Lys Glu Pro Arg Arg
 35 40 45
 Phe Ala Ser Glu Met Lys Gln Cys Val Arg Leu Thr Val His Pro Asn
 50 55 60
 Asn Ile Ser Val Ser Gln Tyr Asn Ala Leu Leu Val Leu Glu Thr Tyr
 65 70 75 80
 Asn Val Pro Glu Leu Ser Ala Gly Val Asn Cys Thr Phe Glu Asp Leu
 85 90 95
 Ser Glu Met Asp Gly Leu Val Val Gly Asn Gln Ile Gln Cys Tyr Ser
 100 105 110
 Pro Ala Ala Lys Glu Val Pro Arg Ile Ile Thr Glu Asn Gly Asp His
 115 120 125
 His Val Val Gln Leu Gln Leu Lys Ser Lys Glu Thr Gly Met Thr Phe
 130 135 140

Ala Ser Thr Ser Phe Val Phe Tyr Asn Cys Ser Val His Asn Ser Cys
 145 150 155 160
 Leu Ser Cys Val Glu Ser Pro Tyr Arg Cys His Trp Cys Lys Tyr Arg
 165 170 175
 His Val Cys Thr Asp Pro Lys Thr Cys Ser Phe Gln Glu Gly Arg Val
 180 185 190
 Lys Leu Pro Glu Val Gly Pro
 195

<210> 9
 <211> 6408
 <212> DNA
 <213> HOMO SAPIEN

<400> 9
 atgcctgctc tgggcccagc tcttctccag gctctctggg ccggggtgggt cctcaccctc 60
 cagcccccctc caccacaatgc attcactccc aatggcacgt atctgcagca cctggcaagg 120
 gaccctacct caggcacctc ctacctgggg gctaccaact tctgttcca gctgagccct 180
 gggctgcagc tggaggccac agtgctccacc ggcctctgtc tagacagcag ggactgcctg 240
 ccacctgtga tgctgatga gtgccccag gccacgcta ccaacaaccc gaatcagctg 300
 ctctgtgtga gcccaggggc cctggtgtga tgcgggagcg tgcaccaggg ggtctgtgaa 360
 cagcgscgco tggggcagct cgagcagctg ctgctgcggc cagagcgggc tggggacaca 420
 caatatgtgg ctgccaatga tctctcggtc agcacggtgg ggctggtagc ccagggcttg 480
 gcaggggagc cctctctgtt tgtggggcga ggatacacca gcagggggtgt ggggggtggg 540
 attccacca tcacaacccg ggccctgtgg ccgcccagacc ccaagctgc ctctcctat 600
 gaggagacag ccaagctggc agtggggcgc ctctccgagt acagccacca ctctgtgagt 660
 gcctttgcac gtggggccag cgcctaactc ctgttctctg ggcgggacct gcaggctcag 720
 tctagagctt tctgtgccta tgtatctcga gtgtgtctcc gggaccagca ctactactcc 780
 tatgtggagt tgcctctggc tgcgaaggt ggccgctacg ggctgatcca gctgcagct 840
 gtggccacyt ccagggaggt ggccgatggg gagggtgctt ttgcagcttt ctctcggct 900
 gcacccccca ctgtgggccc gcccccatcg gggctgtctg gggcatctgg agcctctgcc 960
 ctctgtgcct tccccctgga tgaggtggac cggcttgcta atcgcacgg agatgcctgc 1020
 tacaccggg agggctcgtc tgaggatggg accgaggtgg cctacatcga gtatgatgtc 1080
 aattctgact gtgcacagct gccagtgagc accctggatg ctatccctg tggctcagac 1140
 cacacgccc gccccatggc cagcgggtgc ccgctggaag ccacaccaat tctggagtgg 1200
 ccagggattc agctaaccag tgtggcagtc accatggaag atggacacac cactgccttc 1260
 ctgggtgata gtcaagggca gctgcacagg gtctacttgg gcccagggag cgatggccac 1320
 ccaactacca cacagagcat ccagcagggg tctgcagtga gcagagacct cacctttgat 1380
 gggacccctt agcacctgta tgtcatgacc cagagcacac ttctgaaggt tcctgtggct 1440
 tctctgtctc agcacctgga ctgtgcactc tgccttgctc acagggaccc atactgtggg 1500
 tgggtcgtgc tctctggcag gtgcagtcgc cgttctgagt gctcgagggg ccagggccca 1560
 gagcagtgcc tatggagctt ccagcctgag ctgggctgtc tgcaagtggc agccatgagt 1620
 cctgccaca tcagccgaga ggagacgagg gagggtttcc tatcagtgcc agacctgcca 1680
 cccctgtggc caggggagtc atattcctgc cactttgggg aacatcagag tctgtccctg 1740
 ctgactgggt ctggtgtgat gtgccccctc ccagacccta gtgaggcccc agtgtgccc 1800
 agaggagccg actacgtatc cgtgagcgtg gagctcagat ttggcgctgt tggatcgcc 1860
 aaaacttccc tctctttcta tgactgtgtg gcggtcactg aactccgccc atctgcgcag 1920
 tgccaggcgt gtgtgagcag ccgctggggg tgtaactggg gtgtctggca gcacctgtgc 1980
 acccacaagg cctcgtgtga tgcctggccc atggttgcaa gccatcagag ccgctgtgtc 2040
 tccccagacc ctctctcaag aggttgaccc agccccctcc caccacagc ccccaagcc 2100
 ctggccaccc ctgtctctga cacccttccc gtggagcctg gggctccctc cacagccaca 2160
 gcttcggaga tctcactgct ggctagtctc tcctgtctca gcccttgggg gccatgggca 2220
 ggttctggct ccatatcttc accctggctc acagggctgc ctctccatga ggagccctcc 2280
 cctcccagcc cccaaaatgg acctggaacc gctgtccctg cccccactga cttcagacc 2340
 tcagccacac ctgaggacct ctgtggcctc ccgctgtcac cgtcagaggt agcagcagtg 2400

ccccctgcag	acccttggccc	cgaggctctt	catcccacag	tgcccctgga	cctgcccctt	2460
gccactgttc	ctgccaccac	tttcccaggg	gccatgggct	ccgtgaagcc	cgccctggac	2520
tgggtccacga	gagaaggcgg	cgagctgccc	gaggcgagac	agtggacggg	gggtgacgca	2580
cccgctctct	ccactccac	ggtcctctca	ggtgatggag	actcagcaga	gcttgagggc	2640
ccctcccgccc	ccctcatcct	cccgtccagc	ctcgactacc	agtatgacac	ccccgggctc	2700
tgggaagctgg	aagaggcgac	cttgggggca	agctcctgcc	cctgtgtgga	gagcgttcag	2760
ggctccacgt	tgatgcgggt	ccatgtggag	cggaataacc	ggctgctagg	caggaaacctg	2820
caccttttcc	aggatggccc	aggagacaat	gagtgtgtga	tggagctgga	gggctctgag	2880
gtgggtggctg	aggcccgggt	cgagtgtgag	ccacctccag	ataccagtg	ccatgtccacc	2940
tgccagcagc	accagctcag	ctatgaggct	ctgcagccgg	agctccgtgt	ggggctgttt	3000
ctgcgtcggg	ccggccgtct	gcgtgtggac	agtgtcgagg	ggctgcacgt	ggtactgtat	3060
gactgttccg	tgggacatgg	agactgcagc	cgctgccaaa	ctgccatgcc	ccagtatggc	3120
tgtgtgtggt	gtgaggggga	gcgtccacgt	tgtgtgaccc	gggaggcctg	tgggtaggct	3180
gaggctgttg	ccaccagtg	cccagcgccc	ctcatccact	cggtggagcc	actgactggg	3240
ctgttagagc	gaggcacccc	tgtcaccatc	aggggctcca	acctgggcca	gcattgtgag	3300
gatgtgctgc	gcattgttcaa	gtatcccttg	gtcccctgtg	ctgtggatgc	ccaggagtac	3360
gaggtctcca	gcagcctcgt	gtgcatcacc	ggggccagtg	gggaggaggt	ggccggcgcc	3420
acagcgttgg	agggtgccggg	aagaggacgt	ggtgtctcag	aacacgactt	tgccctaccag	3480
gattccgaagg	tccattccat	cttcccggcc	cgccggccca	gagctggggg	cacccgtctc	3540
accctgaatg	gtctcaaagt	ctctactggg	cggtggagg	acatccagtg	ggtgttggga	3600
gaccagcctt	gtcacttgct	gccggagcag	cagtcaaac	aactgcgggt	tgaaccacgc	3660
ccatgcccc	cgctgccc	gtctcctgtg	gtctgtgtgt	tgggggccc	ggaggcggag	3720
cttcaacygc	gcaggttcaa	gtataccctg	gaccccaca	tcacctctgc	tggcccacc	3780
aagagcttcc	tcatgtggag	acgtgagata	tgcgtccgtg	gccagaatct	ggagcttgta	3840
caagacccaa	gaatccgggt	gaccgttgct	tcgagaatgc	tgcagcccaa	ccaggggctt	3900
ggacggaggc	gtgcgctggt	cccggagacg	gcattgtccc	tggaccctc	ctgcagtagc	3960
cagcaatttg	aggagccgtg	ccatgtcaac	tcctcccagc	tcacacagtg	ccgcacacct	4020
gccttccacg	gccttgcctga	ggaccctctg	gtccgggttg	aatttatcct	tgacaacctg	4080
gtctttgact	ttgcaacact	gaaccccaca	cttttctcct	atgaggccga	ccccacctg	4140
cagccactca	accctgagga	ccccaccatg	ccattccggc	acaagcctgg	gagtggttcc	4200
tccgtggagg	gggagaacct	ggaccttgca	atgtccaagg	aggagggtgt	ggctatgata	4260
ggggatggcc	cctgtgtggt	gaagacgctg	acgcggcacc	acctgtactg	cgagcccccc	4320
gtggagcagc	ccctgccacg	gcaccatgcc	ctccgagagg	cacctgactc	tttgcttgag	4380
ttcacggtgc	agatggggaa	tcctcgcttc	tcctgggttc	acgtgacgta	tgacggcgag	4440
agccctgggg	cttttctctg	ggcagcccag	gtgggcttgg	gggtgggcac	ctctctctg	4500
gctctgggtg	tcattcctat	tgtcctcatg	tacaggagga	agagcaagga	ggccctgagg	4560
gactataaga	aggttcagat	ccagctggag	aatctggaga	gcagtgtgcy	ggacctgctc	4620
aagaaggaat	tcacagacct	catgactgag	atgaccgatc	tcaccagtga	cctcttgggc	4680
agcggcatcc	ccttccctga	ctacaagggt	tatgcggaga	ggatcttctt	ccctgggcac	4740
cgcgagtgcg	ccttgccacg	ggacctgggt	gtgcctgaga	gcagacggcc	cactgtagag	4800
caagggtcgg	ggcagctctc	taacctgtct	aacagcaagc	tcctcctcac	caagtccatc	4860
cacacgctgg	agacccagcg	caccttttca	gctcgggacc	gtgcctacgt	ggcatctctg	4920
ctcacctgtg	cactgcattg	gaagcttgag	tatttcaact	acatcctccg	cactctgctc	4980
agtgcactgg	tgtcccagta	tgtggccaag	aaccccaagc	tgatgtctgc	caggacagag	5040
acgtgtgttg	agaagctgct	caccaactgg	atgtccatct	gtctgtatgc	cttcgtgagg	5100
gactccgtag	gggagcctct	gtacatgtct	tttcgaggga	ttaagcacca	agtggaataag	5160
gggccagtgg	acagtgtgac	aggcaaggcc	aaatacacct	tgaacgacaa	ccgcctgctc	5220
agagaggatg	tggagtaccg	tcacctgacc	ttgaatgcac	tattggctgt	ggggcctggg	5280
gcaggagagg	cccaggcgct	gcccgtgaag	gtcctagact	gtgacaccat	ctcccaggca	5340
aaggaagaaga	tgttggacca	gctttataaa	ggagtgcctc	tcacccagcg	gccagacctc	5400
cgcacctctg	atgttgagtg	gcggtctggg	gtggccgggc	acctcattct	ttctgacgag	5460
gatgtcactt	ctgagggtcca	gggtctgttg	agcgccctga	acacactgca	gcattacaag	5520
gtcccagatg	gagcaactgt	ggccctctgc	ccttgcctca	ccaagcatgt	gctccgggaa	5580
aaccaggatt	atgtccctgg	agagcggacc	ccaatgctgg	aggatgtaga	tgaggggggc	5640
atccggccct	ggcacctggg	gaagccaagt	gatgagccgg	agccgcccag	gctccggagg	5700

ggcagccttc	ggggcgggga	gcgtgagcgc	gccaaaggcca	tccttgagat	ctacctgacc	5760
cgcctgctgt	ccatgaagg	caccctgcag	aagttcgtgg	atgacctgtt	ccaggtgatt	5820
ctcagaccca	gccgcgccgt	gccgctcgct	gtgaagtact	tccttgacct	gctggatgag	5880
caggccccagc	agcatggcat	ctccgaccag	gacaccatcc	acatctggaa	gaccaacagc	5940
ttgctcttga	ggttctggat	caatataata	aaaaaccgcg	agtttgtgtt	cgacgtgcaa	6000
acatctgata	acatggatgc	ggtgctcctt	gtcattgcac	agaccttcac	ggacgcctgc	6060
accctggccg	accacaagct	gggcccgggac	tccccgatca	acaaacttct	gtatgcacgg	6120
gcatttcccc	ggtacaagcg	gatggtggaa	aggtagctatg	cagacatcag	acagactgtc	6180
ccagccagcg	accaagagat	gaactctgtc	ctggctgaac	tgctcctggaa	ctactccgga	6240
gacctcgggg	cgcgagtggc	cctgcataaa	ctctacaagt	acatcaacaa	gtactatgac	6300
cagatcatca	ctgccctgga	ggaggatggc	acggcccaga	agatgcagct	gggctatcgg	6360
ctccagcaga	ttgcagctgc	tgtggaaaac	aaggtcacag	atctatag		6408

<210> 10

<211> 2135

<212> PRT

<213> HOMO SAPIEN

<400> 10

Met	Pro	Ala	Leu	Gly	Pro	Ala	Leu	Leu	Gln	Ala	Leu	Trp	Ala	Gly	Trp
1				5					10					15	
Val	Leu	Thr	Leu	Gln	Pro	Leu	Pro	Pro	Thr	Ala	Phe	Thr	Pro	Asn	Gly
			20					25					30		
Thr	Tyr	Leu	Gln	His	Leu	Ala	Arg	Asp	Pro	Thr	Ser	Gly	Thr	Leu	Tyr
			35				40					45			
Leu	Gly	Ala	Thr	Asn	Phe	Leu	Phe	Gln	Leu	Ser	Pro	Gly	Leu	Gln	Leu
	50					55					60				
Glu	Ala	Thr	Val	Ser	Thr	Gly	Pro	Val	Leu	Asp	Ser	Arg	Asp	Cys	Leu
	65				70					75				80	
Pro	Pro	Val	Met	Pro	Asp	Glu	Cys	Pro	Gln	Ala	Gln	Pro	Thr	Asn	Asn
			85					90						95	
Pro	Asn	Gln	Leu	Leu	Leu	Val	Ser	Pro	Gly	Ala	Leu	Val	Val	Cys	Gly
			100					105					110		
Ser	Val	His	Gln	Gly	Val	Cys	Glu	Gln	Arg	Arg	Leu	Gly	Gln	Leu	Glu
		115				120					125				
Gln	Leu	Leu	Arg	Pro	Glu	Arg	Pro	Gly	Asp	Thr	Gln	Tyr	Val	Ala	
	130				135					140					
Ala	Asn	Asp	Pro	Ala	Val	Ser	Thr	Val	Gly	Leu	Val	Ala	Gln	Gly	Leu
	145				150					155				160	
Ala	Gly	Glu	Pro	Leu	Leu	Phe	Val	Gly	Arg	Gly	Tyr	Thr	Ser	Arg	Gly
			165					170						175	
Val	Gly	Gly	Gly	Ile	Pro	Pro	Ile	Thr	Thr	Arg	Ala	Leu	Trp	Pro	Pro
			180					185					190		
Asp	Pro	Gln	Ala	Ala	Phe	Ser	Tyr	Glu	Glu	Thr	Ala	Lys	Leu	Ala	Val
		195					200					205			
Gly	Arg	Leu	Ser	Glu	Tyr	Ser	His	Phe	Val	Ser	Ala	Phe	Ala	Arg	
	210					215				220					
Gly	Ala	Ser	Ala	Tyr	Phe	Leu	Phe	Leu	Arg	Arg	Asp	Leu	Gln	Ala	Gln
	225				230				235					240	
Ser	Arg	Ala	Phe	Arg	Ala	Tyr	Val	Ser	Arg	Val	Cys	Leu	Arg	Asp	Gln
			245					250					255		
His	Tyr	Tyr	Ser	Tyr	Val	Glu	Leu	Pro	Leu	Ala	Cys	Glu	Gly	Gly	Arg
			260				265						270		

Tyr Gly Leu Ile Gln Ala Ala Ala Val Ala Thr Ser Arg Glu Val Ala
 275 280 285
 His Gly Glu Val Leu Phe Ala Ala Phe Ser Ser Ala Ala Pro Pro Thr
 290 295 300
 Val Gly Arg Pro Pro Ser Ala Ala Gly Ala Ser Gly Ala Ser Ala
 305 310 315 320
 Leu Cys Ala Phe Pro Leu Asp Glu Val Asp Arg Leu Ala Asn Arg Thr
 325 330 335
 Arg Asp Ala Cys Tyr Thr Arg Glu Gly Arg Ala Glu Asp Gly Thr Glu
 340 345 350
 Val Ala Tyr Ile Glu Tyr Asp Val Asn Ser Asp Cys Ala Gln Leu Pro
 355 360 365
 Val Asp Thr Leu Asp Ala Tyr Pro Cys Gly Ser Asp His Thr Pro Ser
 370 375 380
 Pro Met Ala Ser Arg Val Pro Leu Glu Ala Thr Pro Ile Leu Glu Trp
 385 390 395 400
 Pro Gly Ile Gln Leu Thr Ala Val Ala Val Thr Met Glu Asp Gly His
 405 410 415
 Thr Ile Ala Phe Leu Gly Asp Ser Gln Gly Gln Leu His Arg Val Tyr
 420 425 430
 Leu Gly Pro Gly Ser Asp Gly His Pro Tyr Ser Thr Gln Ser Ile Gln
 435 440 445
 Gln Gly Ser Ala Val Ser Arg Asp Leu Thr Phe Asp Gly Thr Phe Glu
 450 455 460
 His Leu Tyr Val Met Thr Gln Ser Thr Leu Leu Lys Val Pro Val Ala
 465 470 475 480
 Ser Cys Ala Gln His Leu Asp Cys Ala Ser Cys Leu Ala His Arg Asp
 485 490 495
 Pro Tyr Cys Gly Trp Cys Val Leu Leu Gly Arg Cys Ser Arg Arg Ser
 500 505 510
 Glu Cys Ser Arg Gly Gln Gly Pro Glu Gln Trp Leu Trp Ser Phe Gln
 515 520 525
 Pro Glu Leu Gly Cys Leu Gln Val Ala Ala Met Ser Pro Ala Asn Ile
 530 535 540
 Ser Arg Glu Glu Thr Arg Glu Val Phe Leu Ser Val Pro Asp Leu Pro
 545 550 555 560
 Pro Leu Trp Pro Gly Glu Ser Tyr Ser Cys His Phe Gly Glu His Gln
 565 570 575
 Ser Pro Ala Leu Leu Thr Gly Ser Gly Val Met Cys Pro Ser Pro Asp
 580 585 590
 Pro Ser Glu Ala Pro Val Leu Pro Arg Gly Ala Asp Tyr Val Ser Val
 595 600 605
 Ser Val Glu Leu Arg Phe Gly Ala Val Val Ile Ala Lys Thr Ser Leu
 610 615 620
 Ser Phe Tyr Asp Cys Val Ala Val Thr Glu Leu Arg Pro Ser Ala Gln
 625 630 635 640
 Cys Gln Ala Cys Val Ser Ser Arg Trp Gly Cys Asn Trp Cys Val Trp
 645 650 655
 Gln His Leu Cys Thr His Lys Ala Ser Cys Asp Ala Gly Pro Met Val
 660 665 670
 Ala Ser His Gln Ser Pro Leu Val Ser Pro Asp Pro Pro Ala Arg Gly
 675 680 685
 Gly Pro Ser Pro Ser Pro Thr Ala Pro Lys Ala Leu Ala Thr Pro
 690 695 700
 Ala Pro Asp Thr Leu Pro Val Glu Pro Gly Ala Pro Ser Thr Ala Thr

705					710					715				720
Ala	Ser	Asp	Ile	Ser	Pro	Gly	Ala	Ser	Pro	Ser	Leu	Leu	Ser	Pro
				725						730				735
Gly	Pro	Trp	Ala	Gly	Ser	Gly	Ser	Ile	Ser	Ser	Pro	Gly	Ser	Thr
			740					745					750	
Ser	Pro	Leu	His	Glu	Glu	Pro	Ser	Pro	Pro	Ser	Pro	Gln	Asn	Gly
		755					760					765		
Gly	Thr	Ala	Val	Pro	Ala	Pro	Thr	Asp	Phe	Arg	Pro	Ser	Ala	Thr
		770				775					780			
Glu	Asp	Leu	Leu	Ala	Ser	Pro	Leu	Ser	Pro	Ser	Glu	Val	Ala	Ala
		785			790					795				800
Pro	Pro	Ala	Asp	Pro	Gly	Pro	Glu	Ala	Leu	His	Pro	Thr	Val	Pro
				805					810					815
Asp	Leu	Pro	Pro	Ala	Thr	Val	Pro	Ala	Thr	Thr	Phe	Pro	Gly	Ala
				820					825				830	Met
Gly	Ser	Val	Lys	Pro	Ala	Leu	Asp	Trp	Leu	Thr	Arg	Glu	Gly	Gly
		835					840					845		
Leu	Pro	Glu	Ala	Asp	Glu	Trp	Thr	Gly	Gly	Asp	Ala	Pro	Ala	Phe
		850				855					860			
Thr	Ser	Thr	Leu	Leu	Ser	Gly	Asp	Gly	Asp	Ser	Ala	Glu	Leu	Gly
		865			870				875					880
Pro	Pro	Ala	Pro	Leu	Ile	Leu	Pro	Ser	Ser	Leu	Asp	Tyr	Gln	Tyr
				885					890					895
Thr	Pro	Gly	Leu	Trp	Glu	Leu	Glu	Glu	Ala	Thr	Leu	Gly	Ala	Ser
			900					905					910	
Cys	Pro	Cys	Val	Glu	Ser	Val	Gln	Gly	Ser	Thr	Leu	Met	Pro	Val
			915				920					925		
Val	Glu	Arg	Glu	Ile	Arg	Leu	Gly	Arg	Asn	Leu	His	Leu	Phe	Gln
		930				935						940		
Asp	Gly	Pro	Gly	Asp	Asn	Glu	Cys	Val	Met	Glu	Leu	Glu	Gly	Leu
		945			950				955					960
Val	Val	Val	Glu	Ala	Arg	Val	Glu	Cys	Glu	Pro	Pro	Pro	Asp	Thr
			965						970					975
Cys	His	Val	Thr	Cys	Gln	Gln	His	Gln	Leu	Ser	Tyr	Glu	Ala	Leu
			980					985					990	
Pro	Glu	Leu	Arg	Val	Gly	Leu	Phe	Leu	Arg	Arg	Ala	Gly	Arg	Leu
		995					1000					1005		
Val	Asp	Ser	Ala	Glu	Gly	Leu	His	Val	Val	Leu	Tyr	Asp	Cys	Ser
			1010			1015					1020			
Gly	His	Gly	Asp	Cys	Ser	Arg	Cys	Gln	Thr	Ala	Met	Pro	Gln	Tyr
		1025			1030				1035					1040
Cys	Val	Trp	Cys	Glu	Gly	Glu	Arg	Pro	Arg	Cys	Val	Thr	Arg	Glu
			1045					1050						1055
Cys	Gly	Glu	Ala	Glu	Ala	Val	Ala	Thr	Gln	Cys	Pro	Ala	Pro	Leu
			1060					1065					1070	
His	Ser	Val	Glu	Pro	Leu	Thr	Gly	Pro	Val	Asp	Gly	Gly	Thr	Arg
		1075					1080					1085		
Thr	Ile	Arg	Gly	Ser	Asn	Leu	Gly	Gln	His	Val	Gln	Asp	Val	Leu
		1090				1095					1100			
Met	Val	Thr	Val	Ala	Gly	Val	Pro	Cys	Ala	Val	Asp	Ala	Gln	Glu
		1105			1110				1115					1120
Glu	Val	Ser	Ser	Ser	Leu	Val	Cys	Ile	Thr	Gly	Ala	Ser	Gly	Glu
			1125					1130						1135
Val	Ala	Gly	Ala	Thr	Ala	Val	Glu	Val	Pro	Gly	Arg	Gly	Arg	Gly
			1140				1145						1150	

Ser Glu His Asp Phe Ala Tyr Gln Asp Pro Lys Val His Ser Ile Phe
 1155 1160 1165
 Pro Ala Arg Gly Pro Arg Ala Gly Gly Thr Arg Leu Thr Leu Asn Gly
 1170 1175 1180
 Ser Lys Leu Leu Thr Gly Arg Leu Glu Asp Ile Arg Val Val Val Gly
 1185 1190 1195 1200
 Asp Gln Pro Cys His Leu Leu Pro Glu Gln Gln Ser Glu Gln Leu Arg
 1205 1210 1215
 Cys Glu Thr Ser Pro Arg Pro Thr Pro Ala Thr Leu Pro Val Ala Val
 1220 1225 1230
 Trp Phe Gly Ala Thr Glu Arg Arg Leu Gln Arg Gly Gln Phe Lys Tyr
 1235 1240 1245
 Thr Leu Asp Pro Asn Ile Thr Ser Ala Gly Pro Thr Lys Ser Phe Leu
 1250 1255 1260
 Ser Gly Gly Arg Glu Ile Cys Val Arg Gly Gln Asn Leu Asp Val Val
 1265 1270 1275 1280
 Gln Thr Pro Arg Ile Arg Val Thr Val Val Ser Arg Met Leu Gln Pro
 1285 1290 1295
 Ser Gln Gly Leu Gly Arg Arg Arg Val Val Pro Glu Thr Ala Cys
 1300 1305 1310
 Ser Leu Gly Pro Ser Cys Ser Ser Gln Gln Phe Glu Glu Pro Cys His
 1315 1320 1325
 Val Asn Ser Ser Gln Leu Ile Thr Cys Arg Thr Pro Ala Leu Pro Gly
 1330 1335 1340
 Leu Pro Glu Asp Pro Trp Val Arg Val Glu Phe Ile Leu Asp Asn Leu
 1345 1350 1355 1360
 Val Phe Asp Phe Ala Thr Leu Asn Pro Thr Pro Phe Ser Tyr Glu Ala
 1365 1370 1375
 Asp Pro Thr Leu Gln Pro Leu Asn Pro Glu Asp Pro Thr Met Pro Phe
 1380 1385 1390
 Arg His Lys Pro Gly Ser Val Phe Ser Val Glu Gly Glu Asn Leu Asp
 1395 1400 1405
 Leu Ala Met Ser Lys Glu Glu Val Val Ala Met Ile Gly Asp Gly Pro
 1410 1415 1420
 Cys Val Val Lys Thr Leu Thr Arg His His Leu Tyr Cys Glu Pro Pro
 1425 1430 1435 1440
 Val Glu Gln Pro Leu Pro Arg His His Ala Leu Arg Glu Ala Pro Asp
 1445 1450 1455
 Ser Leu Pro Glu Phe Thr Val Gln Met Gly Asn Leu Arg Phe Ser Leu
 1460 1465 1470
 Gly His Val Gln Tyr Asp Gly Glu Ser Pro Gly Ala Phe Pro Val Ala
 1475 1480 1485
 Ala Gln Val Gly Leu Gly Val Gly Thr Ser Leu Leu Ala Leu Gly Val
 1490 1495 1500
 Ile Ile Ile Val Leu Met Tyr Arg Arg Lys Ser Lys Gln Ala Leu Arg
 1505 1510 1515 1520
 Asp Tyr Lys Lys Val Gln Ile Gln Leu Glu Asn Leu Glu Ser Ser Val
 1525 1530 1535
 Arg Asp Arg Cys Lys Lys Glu Phe Thr Asp Leu Met Thr Glu Met Thr
 1540 1545 1550
 Asp Leu Thr Ser Asp Leu Leu Gly Ser Gly Ile Pro Phe Leu Asp Tyr
 1555 1560 1565
 Lys Val Tyr Ala Glu Arg Ile Phe Phe Pro Gly His Arg Glu Ser Pro
 1570 1575 1580
 Leu His Arg Asp Leu Gly Val Pro Glu Ser Arg Arg Pro Thr Val Glu

1585	Gln	Gly	Leu	Gly	Gln	Leu	Ser	Asn	Leu	Leu	Asn	Ser	Lys	Leu	Phe	Leu	1600
	Thr	Lys	Phe	Ile	His	Thr	Leu	Glu	Ser	Gln	Arg	Thr	Phe	Ser	Ala	Arg	1615
	Asp	Arg	Ala	Tyr	Val	Ala	Ser	Leu	Leu	Thr	Val	Ala	Leu	His	Gly	Lys	1630
	Leu	Glu	Tyr	Phe	Thr	Asp	Ile	Leu	Arg	Thr	Leu	Leu	Ser	Asp	Leu	Val	1645
	Ala	Gln	Tyr	Val	Ala	Lys	Asn	Pro	Lys	Leu	Met	Leu	Arg	Arg	Thr	Glu	1660
	Thr	Val	Val	Glu	Lys	Leu	Leu	Thr	Asn	Trp	Met	Ser	Ile	Cys	Leu	Tyr	1675
	Thr	Phe	Val	Arg	Asp	Ser	Val	Gly	Glu	Pro	Leu	Tyr	Met	Leu	Phe	Arg	1690
	Gly	Ile	Lys	His	Gln	Val	Asp	Lys	Gly	Pro	Val	Asp	Ser	Val	Thr	Gly	1705
	Lys	Ala	Lys	Tyr	Thr	Leu	Asn	Asp	Asn	Arg	Leu	Leu	Arg	Glu	Asp	Val	1720
	Glu	Tyr	Arg	Pro	Leu	Thr	Leu	Asn	Ala	Leu	Leu	Ala	Val	Gly	Pro	Gly	1735
	Ala	Gly	Glu	Ala	Gln	Gly	Val	Pro	Val	Lys	Val	Leu	Asp	Cys	Asp	Thr	1750
	Ile	Ser	Gln	Ala	Lys	Glu	Lys	Met	Leu	Asp	Gln	Leu	Tyr	Lys	Gly	Val	1765
	Pro	Leu	Thr	Gln	Arg	Pro	Asp	Pro	Arg	Thr	Leu	Asp	Val	Glu	Trp	Arg	1780
	Ser	Gly	Val	Ala	Gly	His	Leu	Ile	Leu	Ser	Asp	Glu	Asp	Val	Thr	Ser	1795
	Glu	Val	Gln	Gly	Leu	Trp	Arg	Arg	Leu	Asn	Thr	Leu	Gln	His	Tyr	Lys	1810
	Val	Pro	Asp	Gly	Ala	Thr	Val	Ala	Leu	Val	Pro	Cys	Leu	Thr	Lys	His	1825
	Val	Leu	Arg	Glu	Asn	Gln	Asp	Tyr	Val	Pro	Gly	Glu	Arg	Thr	Pro	Met	1840
	Leu	Glu	Asp	Val	Asp	Glu	Gly	Gly	Ile	Arg	Pro	Trp	His	Leu	Val	Lys	1855
	Pro	Ser	Asp	Glu	Pro	Glu	Pro	Pro	Arg	Pro	Arg	Arg	Gly	Ser	Leu	Arg	1870
	Gly	Gly	Glu	Arg	Glu	Arg	Ala	Lys	Ala	Ile	Pro	Glu	Ile	Tyr	Leu	Thr	1885
	Arg	Leu	Leu	Ser	Met	Lys	Gly	Thr	Leu	Gln	Lys	Phe	Val	Asp	Asp	Leu	1900
	Phe	Gln	Val	Ile	Leu	Ser	Thr	Ser	Arg	Pro	Val	Pro	Leu	Ala	Val	Lys	1915
	Tyr	Phe	Phe	Asp	Leu	Leu	Asp	Glu	Gln	Ala	Gln	Gln	His	Gly	Ile	Ser	1930
	Asp	Gln	Asp	Thr	Ile	His	Ile	Trp	Lys	Thr	Asn	Ser	Leu	Pro	Leu	Arg	1945
	Phe	Trp	Ile	Asn	Ile	Ile	Lys	Asn	Pro	Gln	Phe	Val	Phe	Asp	Val	Gln	1960
	Thr	Ser	Asp	Asn	Met	Asp	Ala	Val	Leu	Leu	Val	Ile	Ala	Gln	Thr	Phe	1975
	Met	Asp	Ala	Cys	Thr	Leu	Ala	Asp	His	Lys	Leu	Gly	Arg	Asp	Ser	Pro	2000

Ile Asn Lys Leu Leu Tyr Ala Arg Asp Ile Pro Arg Tyr Lys Arg Met
 2035 2040 2045
 Val Glu Arg Tyr Tyr Ala Asp Ile Arg Gln Thr Val Pro Ala Ser Asp
 2050 2055 2060
 Gln Glu Met Asn Ser Val Leu Ala Glu Leu Ser Trp Asn Tyr Ser Gly
 2065 2070 2075 2080
 Asp Leu Gly Ala Arg Val Ala Leu His Glu Leu Tyr Lys Tyr Ile Asn
 2085 2090 2095
 Lys Tyr Tyr Asp Gln Ile Ile Thr Ala Leu Glu Glu Asp Gly Thr Ala
 2100 2105 2110
 Gln Lys Met Gln Leu Gly Tyr Arg Leu Gln Gln Ile Ala Ala Ala Val
 2115 2120 2125
 Glu Asn Lys Val Thr Asp Leu
 2130 2135

<210> 11
 <211> 2190
 <212> DNA
 <213> HOMO SAPIEN

<400> 11
 .atgacctgctc tgggcccagc tcttctccag gctctctggg cggggtgggt cctcaccctc 60
 cagcccccttc caccaaactgc attcactccc aatggcacgt atctgcagca cctggcaagg 120
 gacccccacct caggcacctc ctacctgggg gctaccaact tctgtgtcca gctgagccct 180
 gggctgcagc tggagggccac agtgtccacc ggccctgtgc tagacagcag ggactgcctg 240
 ccacctgtga tgctgatga gtgccccag gccacagcta ccaacaacc gaatacagctg 300
 ctctgggtga gccagggggc cctgggtgta tgcgggagcg tgaccagggg ggctgtgtga 360
 cagcggggccc tggggcagct cgagcagctg ctgctgcggc cagagcggcc tggggacaca 420
 caatatgtgg ctgccaatga tcttgcggtc agcacgggtg ggctggtagc ccagggtctg 480
 gcaggggagc cctctctgtt tgtggggcga ggatacaca gcagggtgtg ggggggtggc 540
 attccaccca tcacaacccg ggcctctgtg ccgcccagac ccaagctgc tttctcttat 600
 gaggagacag ccaagctggc agtggggcgc ctctccagat acagccacca ctctgtgagt 660
 gcctttgcac gtggggccag cgctacttct ctgttctcgc ggcgggacct gcaggctcag 720
 tctagagctt tctgtgccta tgtatctcga gtgtgtctcc gggaccagca ctactactcc 780
 tatgtggagt tgctctggc ctgcgaaggt ggccgctacg ggctgatcca ggctgcagct 840
 gtggccacgt ccaggagggt ggcccatggg gaggtgctct ttgcagcttt ctctcggct 900
 gcacccccca ctgtggggcg gcccccacgc gcggctgtct gggcatctgg agcctctcgc 960
 ctctgtgctt tccccttga tgaggtggac cggcttgcta atcgacgcg agatgcctgc 1020
 tacacccggg agggctctgc tgaggtggg accgaggtgg cctacatcga gtatgatgtc 1080
 aattctgact gtgcacagct gccagtgagc accctggatg cttatccctg tggctcagac 1140
 cacacgcca gccccatggc cagccgggtc ccgctggaag ccacaccaat tctggagtgg 1200
 ccagggtatc agctaacagc tgtggcagtc accatggaag atggacacac catcgctttc 1260
 ctgggtgata gctcaaggga gctgcacagg gtctacttgg gccaggggag cgatggccac 1320
 ccatactcca cacagagcat ccagcagggg tctgcagtga gcagagacct cactcttgat 1380
 gggacctttg agcacctgta tgtcatgacc cagagcacac ttctgaaggt tctgtggct 1440
 tctgtgtctc agcacctgga ctgtgcattc tgcttctcgc acagggaccc atactgtggg 1500
 ttgtgtgtgc tcttggcag gtgcagtcgc cgttctgagt gctcaggggg ccaggggcca 1560
 gagcagtggc tatggagctt ccagccctgag ctgggctgtc tgcaagtggc agccatgagt 1620
 cctgccaca tcagccgaga ggagacgagg gaggttttcc tatcagtgc agacctgcca 1680
 cccctgtggc cagggggagtc atattctctg cactttgggg aacatcagag tctgcccctg 1740
 ctgactgtgt ctgggtgtat gtgccccctc ccagacccta gtgagggccc agtgcctgcc 1800
 agaggagccg actacgtatc cgtgagcgtg gagctcagat ttggcgctgt tgtgatcgcc 1860
 aaaacttccc tctctttcta tgactgtgtg gcggtcactg aactccgccc atctgcgcag 1920
 tgccaggcct cctgtgagcag ccgtgtgggg tgtaactggt gtgtctggga gcacctgtgc 1980
 accccaagg gctcgtgtga tgcggggccc atgggtgcaa gccatcaggt gatggagact 2040

cagcagagct tgagggccct cccgcccccc tcatacctccc gtccagcctc gactaccagt 2100
 atgacacccc cgggctctgg gagctggaag aggcgacctt gggggcaagc tcctgcccct 2160
 gtgtggagag cggtcagggc tccacgttga 2190

<210> 12
 <211> 729
 <212> PRT
 <213> HOMO SAPIEN

<400> 12
 Met Pro Ala Leu Gly Pro Ala Leu Leu Gln Ala Leu Trp Ala Gly Trp
 1 5 10 15
 Val Leu Thr Leu Gln Pro Leu Pro Pro Thr Ala Phe Thr Pro Asn Gly
 20 25 30
 Thr Tyr Leu Gln His Leu Ala Arg Asp Pro Thr Ser Gly Thr Leu Tyr
 35 40 45
 Leu Gly Ala Thr Asn Phe Leu Phe Gln Leu Ser Pro Gly Leu Gln Leu
 50 55 60
 Glu Ala Thr Val Ser Thr Gly Pro Val Leu Asp Ser Arg Asp Cys Leu
 65 70 75 80
 Pro Pro Val Met Pro Asp Glu Cys Pro Gln Ala Gln Pro Thr Asn Asn
 85 90 95
 Pro Asn Gln Leu Leu Val Ser Pro Gly Ala Leu Val Val Cys Gly
 100 105 110
 Ser Val His Gln Gly Val Cys Glu Gln Arg Arg Leu Gly Gln Leu Glu
 115 120 125
 Gln Leu Leu Leu Arg Pro Glu Arg Pro Gly Asp Thr Gln Tyr Val Ala
 130 135 140
 Ala Asn Asp Pro Ala Val Ser Thr Val Gly Leu Val Ala Gln Gly Leu
 145 150 155 160
 Ala Gly Glu Pro Leu Phe Val Gly Arg Gly Tyr Thr Ser Arg Gly
 165 170 175
 Val Gly Gly Gly Ile Pro Pro Ile Thr Thr Arg Ala Leu Trp Pro Pro
 180 185 190
 Asp Pro Gln Ala Ala Phe Ser Tyr Glu Glu Thr Ala Lys Leu Ala Val
 195 200 205
 Gly Arg Leu Ser Glu Tyr Ser His His Phe Val Ser Ala Phe Ala Arg
 210 215 220
 Gly Ala Ser Ala Tyr Phe Leu Phe Leu Arg Arg Asp Leu Gln Ala Gln
 225 230 235 240
 Ser Arg Ala Phe Arg Ala Tyr Val Ser Arg Val Cys Leu Arg Asp Gln
 245 250 255
 His Tyr Tyr Ser Tyr Val Glu Leu Pro Leu Ala Cys Glu Gly Gly Arg
 260 265 270
 Tyr Gly Leu Ile Gln Ala Ala Ala Val Ala Thr Ser Arg Glu Val Ala
 275 280 285
 His Gly Glu Val Leu Phe Ala Ala Phe Ser Ser Ala Ala Pro Pro Thr
 290 295 300
 Val Gly Arg Pro Pro Ser Ala Ala Ala Gly Ala Ser Gly Ala Ser Ala
 305 310 315 320
 Leu Cys Ala Phe Pro Leu Asp Glu Val Asp Arg Leu Ala Asn Arg Thr
 325 330 335
 Arg Asp Ala Cys Tyr Thr Arg Glu Gly Arg Ala Glu Asp Gly Thr Glu
 340 345 350
 Val Ala Tyr Ile Glu Tyr Asp Val Asn Ser Asp Cys Ala Gln Leu Pro

```

      355      360      365
Val Asp Thr Leu Asp Ala Tyr Pro Cys Gly Ser Asp His Thr Pro Ser
370      375      380
Pro Met Ala Ser Arg Val Pro Leu Glu Ala Thr Pro Ile Leu Glu Trp
385      390      395
Pro Gly Ile Gln Leu Thr Ala Val Ala Val Thr Met Glu Asp Gly His
405      410      415
Thr Ile Ala Phe Leu Gly Asp Ser Gln Gly Gln Leu His Arg Val Tyr
420      425      430
Leu Gly Pro Gly Ser Asp Gly His Pro Tyr Ser Thr Gln Ser Ile Gln
435      440      445
Gln Gly Ser Ala Val Ser Arg Asp Leu Thr Phe Asp Gly Thr Phe Glu
450      455      460
His Leu Tyr Val Met Thr Gln Ser Thr Leu Leu Lys Val Pro Val Ala
465      470      475
Ser Cys Ala Gln His Leu Asp Cys Ala Ser Cys Leu Ala His Arg Asp
485      490      495
Pro Tyr Cys Gly Trp Cys Val Leu Leu Gly Arg Cys Ser Arg Arg Ser
500      505      510
Glu Cys Ser Arg Gly Gln Gly Pro Glu Gln Trp Leu Trp Ser Phe Gln
515      520      525
-Pro-Glu-Leu-Gly-Cys-Leu-Gln-Val-Ala-Ala-Met-Ser-Pro-Ala-Asn-Ile
530      535      540
Ser Arg Glu Glu Thr Arg Glu Val Phe Leu Ser Val Pro Asp Leu Pro
545      550      555
Pro Leu Trp Pro Gly Glu Ser Tyr Ser Cys His Phe Gly Glu His Gln
565      570      575
Ser Pro Ala Leu Leu Thr Gly Ser Gly Val Met Cys Pro Ser Pro Asp
580      585      590
Pro Ser Glu Ala Pro Val Leu Pro Arg Gly Ala Asp Tyr Val Ser Val
595      600      605
Ser Val Glu Leu Arg Phe Gly Ala Val Val Ile Ala Lys Thr Ser Leu
610      615      620
Ser Phe Tyr Asp Cys Val Ala Val Thr Glu Leu Arg Pro Ser Ala Gln
625      630      635
Cys Gln Ala Cys Val Ser Ser Arg Trp Gly Cys Asn Trp Cys Val Trp
645      650      655
Gln His Leu Cys Thr His Lys Ala Ser Cys Asp Ala Gly Pro Met Val
660      665      670
Ala Ser His Gln Val Met Glu Thr Gln Gln Ser Leu Arg Ala Leu Pro
675      680      685
Pro Pro Ser Ser Ser Arg Pro Ala Ser Thr Thr Ser Met Thr Pro Pro
690      695      700
Gly Ser Gly Ser Trp Lys Arg Arg Pro Trp Gly Gln Ala Pro Ala Pro
705      710      715
Val Trp Arg Ala Phe Arg Ala Pro Arg
725

```


(19) World Intellectual Property Organization
International Bureau



(43) International Publication Date
1 March 2001 (01.03.2001)

PCT

(10) International Publication Number
WO 01/14420 A3

(51) International Patent Classification⁷: C12N 15/12,
15/62, 15/63, C07K 14/705, 16/28, C12P 21/02, A61K
38/17, 39/395, G01N 33/53

the University of California, 12th floor, 1111 Franklin
Street, Oakland, CA 94607-5200 (US). TAMAGNONE,
Luca [IT/IT]; Corso Einaudi, 43, I-10129 Torino (IT).

(21) International Application Number: PCT/US00/23365

(74) Agent: COX, Niki, D.: Biogen, Inc., 14 Cambridge Cen-
ter, Cambridge, MA 02142 (US).

(22) International Filing Date: 25 August 2000 (25.08.2000)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
60/150,576 25 August 1999 (25.08.1999) US

(71) Applicants (for all designated States except US): UNI-
VERSITY OF TORINO [IT/IT]; Department of Biomed-
ical Sciences and Human Oncology, IRCC, SP 142, I-10060
Candiolo (IT). REGENTS OF THE UNIVERSITY OF
CALIFORNIA [US/US]; 12th floor, 1111 Franklin Street,
Oakland, CA 94607-5200 (US).

(81) Designated States (national): AE, AG, AL, AM, AT, AU,
AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ,
DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR,
HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR,
LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ,
NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM,
TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW.

(84) Designated States (regional): ARIPO patent (GH, GM,
KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW), Eurasian
patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European
patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE,
IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG,
CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).

Published:

— with international search report

(72) Inventors; and

(75) Inventors/Applicants (for US only): ARTIGIANI, Ste-
fania [IT/IT]; Corso Brunelleschi, 121/B, I-10100 Torino
(IT). COMOGLIO, Paolo, M. [IT/IT]; Strada Valsalice,
183/8, I-10100 Torino (IT). GOODMAN, Corey, S.
[US/US]; Regents of the University of California, 12th
floor, 1111 Franklin Street, Oakland, CA 94607-5200
(US). TESIER-LAVIGNE, Marc [US/US]; Regents of

(88) Date of publication of the international search report:
10 May 2002

For two-letter codes and other abbreviations, refer to the "Guid-
ance Notes on Codes and Abbreviations" appearing at the begin-
ning of each regular issue of the PCT Gazette.

(54) Title: NOVEL MEMBERS OF THE PLEXIN FAMILY AND USES THEREOF

(57) Abstract: The invention provides methods and compositions related to novel plexins. The polypeptides may be produced re-
combinantly from transformed host cells and from the disclosed plexin encoding nucleic acids or purified from human cells. The
invention provides isolated plexin hybridization probes and primers capable of specifically hybridizing with the disclosed plexin
genes, plexin-specific binding agents such as specific antibodies, and methods of making and using the subject compositions in diag-
nosis, therapy and in biopharmaceutical industry. The invention also provides novel plexin neuropilin multimeric receptor complexes
for semaphorins and methods of use thereof, including but not limited to, the treatment and diagnosis of neurological disease and
neuroregeneration, immune modulation, and viral and oncological diseases.



WO 01/14420 A3

II INTERNATIONAL SEARCH REPORT

Int. Application No
PCT/US 00/23365

A. CLASSIFICATION OF SUBJECT MATTER IPC 7 C12N15/12 C12N15/62 C12N15/63 C07K14/705 C07K16/28 C12P21/02 A61K38/17 A61K39/395 G01N33/53		
According to International Patent Classification (IPC) or to both national classification and IPC		
B. FIELDS SEARCHED Minimum documentation searched (classification system followed by classification symbols) IPC 7 C07K		
Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched		
Electronic data base consulted during the international search (name of data base and, where practical, search terms used) EPO-Internal, WPI Data, PAJ, BIOSIS, MEDLINE, EMBASE, SCISEARCH, STRAND		
C. DOCUMENTS CONSIDERED TO BE RELEVANT		
Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	EMBL DATABASE EMHUM4:HSAB2313; ACCESSION-NO: AB002313, 1 July 1997 (1997-07-01), XP002157964	1-5
Y	the whole document & DATABASE SWALL:015031; ACCESSION-NO: 015031, 1 January 1998 (1998-01-01), the whole document & NAGASE, T. ET AL.: "Prediction of the coding sequences of unidentified human genes. VII. The complete sequences of 100 new cDNA clones from brain which can code for large proteins in vitro" DNA RESEARCH, vol. 4, 1997, pages 141-150, XP002102085 page 142 -page 150 'Results and Discussion' figure 3; tables 1,2 ---	6-9
-/-		
<input checked="" type="checkbox"/> Further documents are listed in the continuation of box C. <input checked="" type="checkbox"/> Patent family members are listed in annex.		
* Special categories of cited documents : "A" document defining the general state of the art which is not considered to be of particular relevance "E" earlier document but published on or after the international filing date "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) "O" document referring to an oral disclosure, use, exhibition or other means "P" document published prior to the international filing date but later than the priority date claimed "T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art. "Z" document member of the same patent family		
Date of the actual completion of the international search 6 August 2001		Date of mailing of the international search report 31.08.01
Name and mailing address of the ISA European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016		Authorized officer Donath, C

II INTERNATIONAL SEARCH REPORT

Int. Application No

PCT/US 00/23365

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	EMBL DATABASE EM_HUM:AB014520; ACCESSION-NO.:AB014520, 15 July 1998 (1998-07-15), XP002173834	1-5
Y	the whole document & ISHIKAWA, K.-I. ET AL.: "Prediction of the coding sequences of unidentified human genes. X. The complete sequences of 100 new cDNA clones from brain which can code for large proteins in vitro" DNA RESEARCH, vol. 5, 30 June 1998 (1998-06-30), pages 169-176, XP002121149 page 172 -page 176 * "Results and Discussion" * figures 1,2; tables 1-3	6-9
X	EMBL DATABASE EMHUM4:HS5211110; ACCESSION-NO: U52111, 9 May 1996 (1996-05-09), XP002173835	1-5
Y	page 12 -page 13 * Gene="PLXB3" and product="plexin-related protein" *	6-9
X	EMBL DATABASE EMHUM6:HSOCTPROT; ACCESSION-NO: X87831, 6 February 1996 (1996-02-06), XP002173836 the whole document	1-3
X	EMBL DATABASE EM_OV:XLPLEX; ACCESSION-NO:D38175, 25 August 1995 (1995-08-25), XP002173837 the whole document & OHTA, K. ET AL.: "Plexin: a novel neuronal cell surface molecule that mediates cell adhesion via a homophilic binding mechanism in the presence of calcium ions" NEURON, vol. 14, 1995, pages 1189-1199, XP001013227	1-3
X	WO 99 04263 A (THE JOHN HOPKINS UNIVERSITY SCHOOL OF MEDICINE) 28 January 1999 (1999-01-28)	10,11
Y	page 5, line 9 -page 10, line 16 page 16, line 6 -page 22, line 3 page 23, line 22 -page 24, line 18 page 29, line 6 -page 32, line 4	12,13

-/-

INTERNATIONAL SEARCH REPORT

International Application No.

PCT/US 00/23365

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	KAMEYAMA, T. ET AL.: "Identification of a cell surface protein plexin (the B2) in mouse, and its expression in developing nervous systems" NEUROSCIENCE RESEARCH SUPPLEMENT, vol. 18, 1993, page S115 XP000945106 the whole document	6-9
Y	COMEAU, M. ET AL.: "A poxvirus-encoded semaphorin induces cytokine production from monocytes and binds to a novel cellular semaphorin receptor, VESPR" IMMUNITY, vol. 8, April 1998 (1998-04), pages 473-482, XP000945259 cited in the application page 478 -page 480 'Discussion'	12,13
A	MAESTRINI, E. ET AL.: "A family of transmembrane proteins with homology to the MET-hepatocyte growth factor receptor" PROC.NATL.ACAD.SCI.USA, vol. 93, no. 2, 1996, pages 674-678, XP000941746 the whole document	1-9
P,X	TAMAGNONE, L. ET AL.: "Plexins are a large family of receptors for transmembrane, secreted, and GPI-anchored semaphorins in vertebrates" CELL, vol. 99, 1 October 1999 (1999-10-01), pages 71-80, XP000941702 page 72 -page 78	1-5
P,Y	'Results' and 'Discussion'	6-9,12, 13
P,Y	TAKAHASHI, T. ET AL.: "Plexin-Neuropilin-1 complexes form functional semaphorin-3A receptors" CELL, vol. 99, 1 October 1999 (1999-10-01), pages 59-69, XP000941701 page 60 -page 67 'Results' and 'Discussion'	12,13
A	NAKAMURA, F. ET AL.: "Neuropilin-1 extracellular domains mediate semaphorin D/III-induced growth cone collapse" NEURON, vol. 21, November 1998 (1998-11), pages 1093-1100, XP002174004 cited in the application the whole document	10-13

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 00/23365

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:
see FURTHER INFORMATION sheet PCT/ISA/210
2. ☒ Claims Nos.: 10,11 (partially)
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
see FURTHER INFORMATION sheet PCT/ISA/210
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this International application, as follows:

see additional sheet

1. ☒ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☒ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA 210

Continuation of Box I.1

Although claims 10-13 are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition. Although claim 14 is directed to a diagnostic method practised on the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

Continuation of Box I.2

Claims-Nos.: 10,11 (partially)

Claims 10 and 11 concern a methods which comprise the administration of an agent capable of interfering with the association between a plexin and a neuropilin. Since in the specification this agent is exemplified only to be an antibody raised against the plexin and since it is completely unclear which kind of substances besides said antibody also will be capable of interfering with the association between a plexin and a neuropilin, the scope of said claims is totally ambiguous and undefined as far as any kind of substance other than an antibody raised against the plexin is concerned.

Therefore, the search in respect of claims 10 and 11 has been limited to methods comprising the administration of an antibody raised against the plexin and which is capable of interfering with the association between a plexin and a neuropilin.

The applicant's attention is drawn to the fact that claims, or parts of claims, relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure.

FURTHER INFORMATION CONTINUED FROM PCT/SA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. Claims: 1-9,12-14 (partially)

Claims 1-9,12-14 (partially) refer to the isolation and cloning of a member of the plexin family, plexin B-2. Antibodies specifically binding to this polypeptide, a fusion protein, and methods of diagnosing for tumors, treating a disorder involving aberrant immune regulation involving a signal pathway between plexin and a neuropilin, ~~and suppressing aberrant cell growth, all methods by using either the polypeptide or the antibodies directed against the plexin B-2.~~

2. Claims: 1-9,12-14 (partially)

Claims 1-9,12-14 (partially) refer to the isolation and cloning of a member of the plexin family, plexin B-3. Antibodies specifically binding to this polypeptide, a fusion protein, and methods of diagnosing for tumors, treating a disorder involving aberrant immune regulation involving a signal pathway between plexin and a neuropilin, and suppressing aberrant cell growth, all methods by using either the polypeptide or the antibodies directed against the plexin B-3.

3. Claims: 1-9,12-14 (partially)

Claims 1-9,12-14 (partially) refer to the isolation and cloning of a member of the plexin family, plexin D-1. Antibodies specifically binding to this polypeptide, a fusion protein, and methods of diagnosing for tumors, treating a disorder involving aberrant immune regulation involving a signal pathway between plexin and a neuropilin, and suppressing aberrant cell growth, all methods by using either the polypeptide or the antibodies directed against the plexin D-1.

4. Claims: 1-9,12-14 (partially)

Claims 1-9,12-14 (partially) refer to the isolation and cloning of a member of the plexin family, plexin A-4. Antibodies specifically binding to this polypeptide, a fusion protein, and methods of diagnosing for tumors, treating a disorder involving aberrant immune regulation involving a signal pathway between plexin and a neuropilin, and suppressing aberrant cell growth, all methods by using either the polypeptide or the antibodies directed against the plexin A-4.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

5. Claims: 10,11

Claims 10 and 11 refer either to a method of suppressing or altering aberrant cell growth involving a signalling pathway between a plexin and a neuropilin in a mammal or to a method of treating, suppressing or altering a disorder involving aberrant immune regulation involving a signalling pathway between a plexin and a neuropilin in a mammal, both methods comprises the administration of an agent in general being capable of interfering with the association between the plexin and neuropilin to said mammal.

II INTERNATIONAL SEARCH REPORT

Information on patent family members

Int. Application No

PCT/US 00/23365

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9904263 A	28-01-1999	AU 8405398 A	10-02-1999